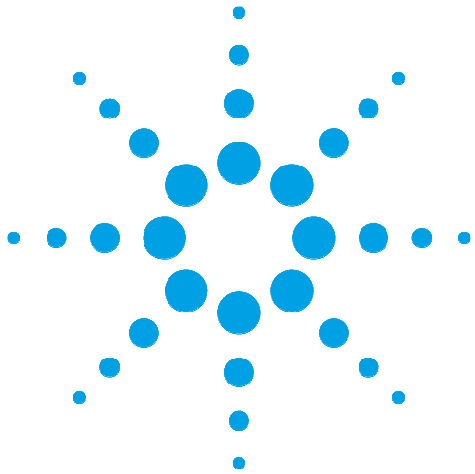

CrysAlis^{Pro}

Data Collection and Processing
Software for X-ray Diffractometers

User Manual

Read the main diffractometer user manual, in particular the Health and Safety information, before operating CrysAlis^{Pro} with the diffractometer.

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1. Introduction

CrysAlis^{Pro} has been designed to provide a user friendly, simple to use, graphical user interface for data collection and data reduction of single crystal X-ray diffraction data.

Based upon the CrysAlis CCD and RED programmes the CrysAlis^{Pro} software provides greatly increased automatic functionality. CrysAlis^{Pro} is a single, multi-threaded programme which combines the functions of the old CCD and RED and can perform each in parallel.

CrysAlis^{Pro} provides direct access to automated data collection and reduction, and the most commonly used functions, in a single multi-threaded programme which can perform each in parallel. Added to this is a command line interface and history window which can be switched on / off and allows access to the complete range of historical and indepth CrysAlis^{Pro} functions.

2. Installation of CrysAlis^{Pro}

To install the CrysAlis^{Pro} software on your diffractometer PC you will need the following programme saved to your computer:

CrysAlisPro171.xx.yy.exe – Where 171.xx.yy is the current version number and the xx and yy values change with each new software update.

You will also need the correction files which are specific to your diffractometer. Without these files the CrysAlis^{Pro} software is not fully functional. To obtain these files copy to your computer the entire 'Corrections' folder from the PC attached to your diffractometer.

1. Double click on the **CrysAlisPro171.xx.yy.exe** programme. Follow the on screen instructions. When asked to select the program elements required for installation click on the box next to **PRO** and **HELP** only (a tick appears in the box) and click on **Next**.
2. Once installation is complete double click on the CrysAlis^{Pro} icon which has been installed on the desktop. The programme may ask you for a set-up file if this is the first installation of CrysAlis^{Pro} on your PC. If required locate the **Corrections** folder which you saved to disk earlier and select the file ***.par**. Click on **OK**.

When installing CrysAlis^{Pro} on a PC remote from your diffractometer, use the process described above. The programme will ask to be pointed towards a data set on launching for the first time, so copy some data onto your remote PC and locate the parameter file (*.par) when prompted.

2.1. Software updates

CrysAlis^{Pro} is continually updated with new features and bug fixes. Major releases incorporating significant new features are available approximately every six months. In between major releases, bug-fixed versions are also available. The latest releases can be downloaded from the Forum website forum.oxford-diffraction.com. Please register on the Forum to access this feature. It is recommended that customers monitor the website and use the latest software.

Each new edition of CrysAlis^{Pro} is released with details of bug fixes and additions since the previous issue. This information can be found in the **Help** menus under **Reference section/Version news**.

CrysAlis^{Pro} users are greatly encouraged to report any software bugs by sending details to XRDSupport@agilent.com. Only by receiving bug reports from experienced users can these be successfully identified and repaired. Ideas for software improvement are also very much welcomed, and all are considered when creating new modules for future editions of CrysAlis^{Pro}.

3. Starting and Closing CrysAlis^{Pro}

3.1. Starting CrysAlis^{Pro}

To start CrysAlis^{Pro} double click on the icon on the windows desktop. When the programme is started on a PC connected to an Agilent diffractometer, CrysAlis^{Pro} will start-up in CCD diffractometer control mode (online) and will include the functionality for control of the CCD, device control and CCD status feedback. On start-up, the software initialises the hardware. Commands cannot be accepted until all hardware initialisation procedures are complete. Wait until the CCD status is shown as ready before issuing commands.

If a second version of CrysAlis^{Pro} is started when a version of CrysAlis^{Pro} is already open and controlling the diffractometer, the second version will open in data reduction/offline mode only (without control of the devices and diffractometer control) and will ask the user to select the 'experiment' that they want to work on from the experiment database. This will also be the case where no diffractometer is connected.

3.2. Closing CrysAlis^{Pro}

To close CrysAlis^{Pro}, click on the X icon in the top right corner of the CrysAlis^{Pro} screen. When in CCD mode, a prompt will appear regarding generator ramping control. For microfocus systems in particular, it is recommended that users turn the generator power down when the diffractometer is not in use. For all other systems it is up to the users individual preferences as to the power settings used when the system is not in use.

4. CrysAlis[Pro] layout and controls

The CrysAlis[Pro] GUI is comprised of a number of different areas. These include:

1. START / STOP button
2. Image control icon bar
3. Information cards
4. Power Tools
5. Diffraction / Image display window
6. Status area
7. Device control

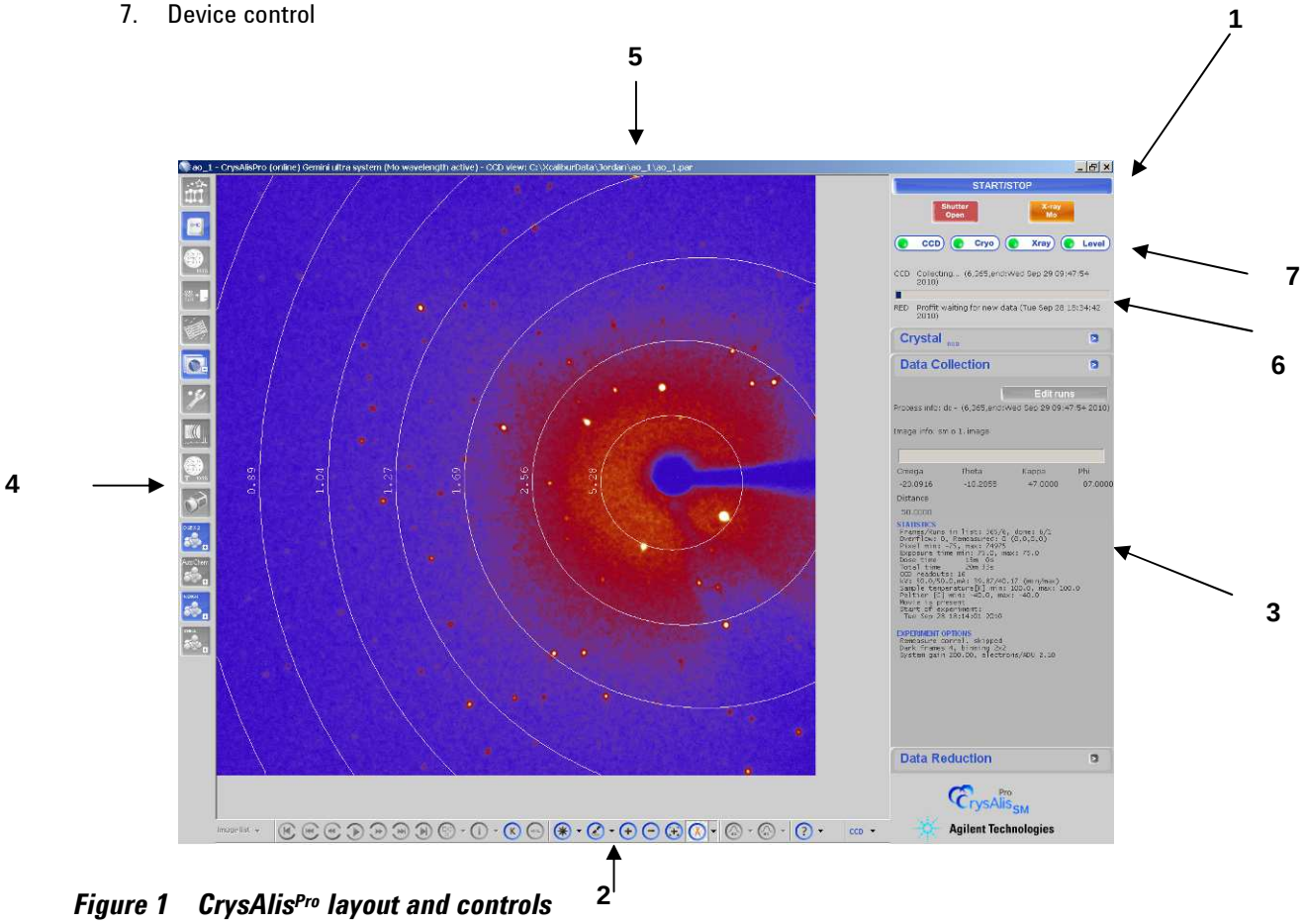


Figure 1 CrysAlis[Pro] layout and controls

4.1. Start / stop

The main control button within the CrysAlis[Pro] GUI is the START / STOP button. The function of this button changes depending upon the current process, but in general it provides access to a range of automatic functions, including:

- Automatic data collection and concurrent automatic data reduction
- Stop All functions

- Start Data reduction only
- Full auto analysis (cell, red)
- Special data collection
- Append special data collection
- Resume data collection/pre-experiment
- Resume all (data collection and reduction)

4.2. Image control icon bar

4.2.1 Key to image control icons

	• Image list
	• Previous run
	• Jump back 10 frames
	• Previous image
	• Play/stop image movie
	• Next image
	• Jump forward 10 images
	• Next run
	• Predictions
	• Pixel / area information
	• Image header information/goniometer angles
	• Find hkl
	• Look up table
	• Colour table
	• Zoom in
	• Zoom out

	• Zoom localiser window
	• Resolution rings
	• 2D Peak profile (a line profile)
	• 3D Peak profile (rocking curve)
	• Help
	• View (CCD/RED/USER)

Figure 2 Key to image control icons

Stepping forward and backward between diffraction images can be controlled by using the image control icons, but also by using the mouse wheel.

4.2.2 Image list

The image list allows the user to open and save diffraction images. Options include: **Open image with explorer**, **Save image** (to save the current diffraction image), **Save image as** (to save the current diffraction image under a new name), **Save jpg** (to save a jpeg graphic image of the currently displayed diffraction image) and **Save bitmap** (to save a bitmap format graphic image of the currently displayed diffraction image). Bitmaps and jpegs can be saved with resolution rings and/or predictions displayed. Zoomed areas of the images can be saved by choosing **Save visible part of image as bitmap or jpeg**.

4.2.3 Predictions

Once the unit cell has been determined, sensible predictions can be overlayed on the diffraction image. Clicking on the predictions icon (in the image control tool bar) switches predictions on and off. Note that these may take a few seconds to display especially when this function is used in conjunction with the movie control buttons such as 'play' where the calculations are all done in advance. If in doubt check the status area in the top right hand corner of the CrysAlis[Pro] GUI.

The predictions overlay a 'cross', 'rhombus or diamond' and 'square' to show positions where the software expects reflections to be if the unit cell is correct. It is therefore used as a visual check of the unit cell and crystal quality. The 'cross' highlights positions where a reflection would be expected to be 'centred' in this frame, the 'rhombus or diamond' shows reflections centred in the previous frame and the 'square' reflections centred in the next frame. (Warning: if a peak search and indexation have not been carried out yet, a default unit cell will be displayed which is obviously not correct.)

4.2.4 Pixel/area information

The pixel /area information icon switches on and off a semi-transparent information display window which appears on the main diffraction window. This window tracks the cursor about the diffraction image highlighting information about the image at the current cursor position. This information includes, the current X and Y pixel position, the Int(ensity) at that position, the H K L indices, the L(orentz), T(heta) and R(esolution).

If the left mouse button is depressed and held a wire box can be drawn around an area / object in the diffraction image window and a semi-transparent window displays the following information about the enclosed area: window start position in pixels, window size end position and dimensions in pixels, maximum and minimum intensity and sigma values for the image and the background.

4.2.5 Header information / K (goniometer angles)

The Header information / K (goniometer angles) icon switches on a window at the top of the diffraction image. This window displays different information depending on whether the computer is offline or online (with goniometer).

When the computer is offline the window displays the current image name and the angles at which it was collected.

When the computer is online the window displays the image name (and its collection angles) and also the current angles of the goniometer. The window provides real-time feedback of what is happening on the machine.

4.2.6 Find hkl

Once you have an indexed unit cell click on the find hkl icon. A dialog box will open. Input the desired H K L in the box at the top of the dialog. Click on **Search**. If the current run list contains this HKL then the frame(s) on which this reflection occur will be listed in the box at the bottom of the dialog. By clicking on a particular line item in this bottom box the main diffraction window will update with the relevant frame image, where the HKL in question is marked with a "+" on the image. If the "+" is difficult to see, use the xy pixel position to help.

4.2.7 Look up table

The look up table affects the main diffraction image display. This is the viewing level above, or intensity threshold above which reflections (or image information) are displayed. Going to a higher look up level displays higher intensity objects and removes lower, background effects from the displayed image.

The look up table can be changed to increasing levels by repeatedly clicking on the icon (which will cycle around to level 1 again after level 13) or by clicking on the arrow to the right of the icon. This will activate a pop-up menu from which the desired look up level can be selected. The currently selected look up table is denoted with a tick mark on the left of the list.

4.2.8 Colour table

The colour table icon allows the user to select from the 11 different colour schemes used to display the diffraction image. Repeated clicking on the colour table icon will cycle through the 11 options or the arrow to the right of the icon can be clicked on to provide a pop-up menu for selection of a particular colour table. The current colour table is highlighted with a tick to the left of the list entry.

4.2.9 Zoom in

Zoom in, magnifies the main diffraction image. Repeated clicking on this icon magnifies the diffraction image in x2 steps to a maximum of x8 magnification. By default the image zooms to the top left hand corner of the image. Use in combination with the zoom localiser.

4.2.10 Zoom out

Zoom out, de-magnifies the main diffraction image. Repeated clicking on this icon de-magnifies the diffraction image in x2 steps to a maximum of x1/8 magnification. Use in combination with the zoom localiser.

4.2.11 Zoom localiser

The zoom localiser is a floating semi-transparent window which displays the full diffraction image (regardless of the zoom used in the main diffraction image). The zoom localiser window can be switched on and off by repeated clicking on the icon in the image control icon bar. Within the zoom localiser window the zoomed area of the main diffraction window is outlined by a wire frame box which can be moved over the zoom localiser window. As the wire frame box is moved over the image the zoomed area in the main diffraction image window is updated. Use in combination with the zoom in and zoom out.

4.2.12 Resolution rings

Resolution rings can be switched on / off by clicking on the button in the image control icon bar. This displays lines of constant 'd' value on the diffraction image to enable a quick visual check of diffraction limits / angular resolution. The default is with resolution rings switched off. Additional settings can be accessed by clicking on the arrow to the right of the resolution ring icon. These settings include the selection of a greater number of rings of constant 'd' value (selectable between the default 3 and the maximum of 12), the display of labels on the rings and the display of rings in black or white.

4.2.13 2D Peak profile

Peak profile allows the user to drag a line through an object in the main diffraction window and obtain a 2D plot of the object. In this way a profile of a reflection (peak) can be obtained to aid the user in the evaluation of the quality of the crystal. Ideal peaks should have a narrow sharp profile with no obvious humps on the side. The profile is displayed in a separate semi-transparent window which defaults to display in the top left corner of the diffraction window.

This window can be resized by dragging out / in the bottom right corner of the window. It can be closed by clicking on the X in the top right corner and it can be moved by holding down the right mouse button with the cursor over the window and moving the mouse. The profile can be saved as a bitmap or jpeg via the floppy disc image in the top right corner of the semi-transparent window. The data can also be saved as a text file or copied to the clipboard by clicking on the arrow symbol.

The semi-transparent window means the profile of peaks can be drawn through the window (i.e. of peaks on the diffraction image behind).

4.2.14 3D Peak profile (Rocking Curve)

This is similar to the 2D peak profile but with the profile taken through consecutive frames. The user needs to define the box size and range around the peak. The box size and range are accessed through the arrows to the right of the icon. The box size needs to be fitted to the size of the reflection. A data file can be saved, or data copied to the clipboard by clicking on the arrow symbol.

4.2.15 Help

The Help icon gives access to online help features, version news and the command reference list.

4.2.16 View

The View icon switches between views. Refer to section 4.6 for more information. Press the view icon repeatedly to cycle through the CCD, RED and USER displays. The header window bar is updated to show which view is being displayed.

In CCD view the images coming off the machine are displayed, but it is not possible to do anything with them when collecting data.

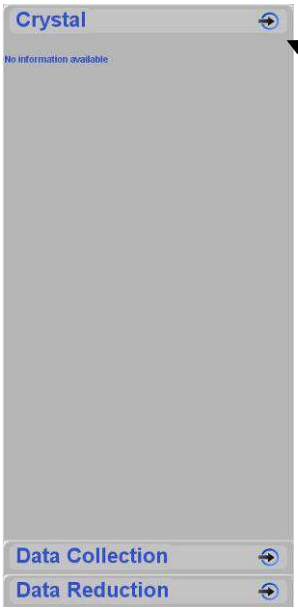
In RED view visual feedback of the data reduction is displayed, but it is not possible to do anything with the images when collecting or reducing data.

In USER view you can read in an individual image, or play through a movie, and perform various analysis tasks whilst data collection and reduction are being performed and updated.

If computer is offline, then only RED and USER are available.

Reference frames (if collected), the beamstop mask, a reciprocal lattice grid overlay for precession images and background subtraction options can be viewed by clicking on the arrow to the right of the CCD/RED/USER icon.

4.3. Information cards



The information cards appear as tabs located on the right-hand side of the screen.

To select a card, click on the text.

The arrow symbols on each information card gives access to several extra features;

Crystal – Edit chemical formula, Edit comment, Show crystal movie, Edit crystal shape, Add experiment performer, Add notes (*.txt) file

Data Collection – Show data collection graphs, Show pre-experiment result

Data Reduction – Show twin log file (if available), Create report (rtf or html)

Figure 3 Information Cards

4.3.1 Pre-Experiment

This information card is only displayed during the automatic pre-experiment and is launched once Start has been clicked on from the pre-experiment set-up dialog.

Displayed on the right hand side of the CrysAlis[Pro] GUI, the pre-experiment window can be minimised by clicking on the 'data collection' information card. Clicking on the pre-experiment header will maximise this window.

The pre-experiment window provides direct feedback of the unit cell, intensity statistics and the defined user set-up for the automatic data collection to follow.

As soon as one frame has been collected the automatic software will attempt to find the unit cell. Once a unit cell has been successfully found it will be displayed in the pre-experiment window, along with information relating to the quality of the cell (e.g. % of reflections indexed), the cell centring and the Laue class. As additional frames are recorded, the unit cell is re-determined and the pre-experiment is updated with the new (or

improved) unit cell. Also displayed are information on the proposed data collection strategy, intensity statistics and a proposed exposure time for the data collection, which are all dependent on the initially specified resolution and I/sig values. These can be altered during the pre-experiment, thereby updating the proposed strategy.

During data collection of the complete experiment, the pre-experiment result can be viewed by clicking on the arrow symbol on the data collection information card. The pre-experiment itself can be re-opened at any time by locating it in the load experiment dialogue window or locating the pre-experiment name.par file in the experiment folder.

4.3.2 Crystal

Following the pre-experiment and during automatic data collection the Crystal information card displays the current unit cell information, the sample chemical formula and z value, and machine model parameters. Also displayed will be the 'Average unit cell from Proffit' (CrysAlis[Pro]'s cell refinement algorithm) and the 'Final unit cell for selected space group'. This is important as, for example, in cases where the data collection strategy is based on a monoclinic cell (with the β angle very close to 90°), it is highly likely that an orthorhombic space group will be chosen and therefore the cell will be transformed as appropriate.

This window can be minimised and maximised as with the pre-experiment information card. Clicking on the arrow symbol at the top of the card gives access to several extra options, including editing of the chemical formula. If the formula is edited here during a data collection, the user must wait for the next cycle of data reduction (this occurs every 25 frames) in order for the changes to take effect. If the formula is changed at any other time, the data must be refinalised to impose the change (see **Chapter 8**).

4.3.3 Data collection

When connected to the diffractometer and collecting data, the data collection information card provides real time progress feedback, displaying the current positions of the omega, theta, kappa, phi axes and the detector distance. Also displayed are a progress bar and the expected end time for the data collection. The end time is updated during the data collection to take into account required frame remeasures, the percentage of which is approximated during the early part of the experiment but then continuously updated.

Information is collected in the data collection tab during data collection. In offline mode this collected information is displayed. For example:

- CCD Peltier maximum and minimum during experiment
- KV and mA maximum and minimum from the generator (if connected)
- Cryojet maximum and minimum temperature
- Overflowed and remeasured reflections
- Maximum and minimum exposure time
- Date, time etc.

4.3.4 Data reduction

The information displayed on this card depends on the data reduction being performed.

The Data Reduction information card displays top level feedback of the progress and results of data reduction and space group determination. The feedback contains information regarding:

- Absorption correction, scaling

- Space group
- Information of face indexed absorption corrections.
- Multi-component information from twin data reduction
- Results (in terms of R_{int}, I/sig, Redundancy, Completeness etc).
- Mosaicity

During the automatic data collection and reduction process this window will only display feedback after the first 25 frames have been collected, since data reduction will not start until this point. This is so that a good estimate of the average background can be established, requiring 25 consecutive frames. The automatic data reduction will process newly collected data in batches of 25 frames and will update the feedback area accordingly, so that by the time the last frames are collected almost all data has already been integrated. This behaviour is the default setting of the auto mode, however, should the user need to re-evaluate the data, they can do so by using the power tools.

4.4. Power tools

4.4.1 Key to power tool icons

	• Lattice Wizard
	• Command line
	• Strategy tool
	• Data finalisation
	• Twin data finalisation
	• CrysAlis^{Pro}
	• Service utility
	• Powder diffraction
	• Multi-Temperature/Wavelength
	• Single images
	• Shell launcher
	• WinGX launcher (if installed)
	• CCP4 launcher (if installed)
	• Olex^{2} launcher (if installed)

	• Crystals launcher (if installed)
	• Jana launcher (if installed)
	• Autochem (if installed)

Figure 4 Key to power tool icons

4.4.2 Lattice Wizard

The lattice wizard icon launches a dialog from which all lattice related functions can be accessed using a GUI. The lattice wizard allows both semi-automatic and manual operation of lattice related functions for when the automatic process fails. The GUI provides for:

- Peak hunting
- Unit cell finding
- Reciprocal space visualisation – using Ewald Explorer
- Indexation with known cell
- Refine instrument model
- Lattice transformation
- Twinning and multi-crystals
- Incommensurates / Quasi-crystals
- Loading and saving your work
- Precession photo reconstruction

As can be seen from the screenshot below the GUI provides buttons for each of the above. Clicking on the buttons such as 'peak hunting' will perform this function automatically using the default settings. If the user wishes to use non-default settings, or change the range of frames etc, then clicking on the arrow to the right of the button offers a greater number of options. Full details regarding use of the lattice wizard can be found in **Chapter 7**.

4.4.3 Command line

This opens a history window and attached command line. From here the user can access all of the existing (historical and new) commands allowing for maximum flexibility and control. The entire experiment and processing can be run completely in this mode or the window can be opened simply to issue specific individual commands. The main part of the window is taken over by a history window, which is always active. This means that even in the fully GUI controlled and automated mode this history window is being updated and can be referred to by the user for further information. When online, using commands which result in image output will display the image in the view corresponding to the command line window tab which is selected. Thus, commands from the RED tab will display images in only the RED view and commands from the CCD tab will display images in only the CCD view. The user has to manually select the proper view to correspond with the selected command line tab. So to control the diffractometer and move the angles to 30 30 30 30 the user would click on the CCD tab, click in the command line and type the 'gt a 30 30 30 30' command. The command line window can be opened or closed at any time and can be resized. An in-depth manual explaining all available commands can be accessed by typing HELP from the command line. Alternatively, descriptions of individual

commands and illustrations of typical usage are available by typing COMMAND HELP in the command line power tool. There is also an options button included which provides access to the machine and software setup.

The command line power tool can be closed by clicking on the 'Close' button located in the bottom right hand corner of the window.

4.4.4 Inspect data collection and reduction results

By opening the **Data finalisation** window, the user can access the results of the automatic data reduction process via five tabbed window panels. The **Data reduction File contents** area contains a summary of cell dimensions and orientation matrix used for integration along with a run by run listing of scan type, angle ranges, exposure time, detector distance and the number of reflections collected in each run.

The **Data reduction output** tab contains the full log of the automatic data reduction process. Starting with the generator settings and data collection temperature, information is provided on any re-measured frames, profile fitting parameters, outlier rejection, scaling, space group determination and final merging statistics, allowing for a thorough inspection of all aspects of the data reduction process.

The **Data collection output** tab gives information obtained during the experiment, specifically relating to remeasured frames and overflows. Any problems encountered during the experiment will also be displayed in this log.

The **Red Graphs** window area displays a run-by-run visual summary of the scaling coefficients (absscale), a frame-by-frame plot of R_{int} and a number of comparisons regarding the coverage and or completeness of the data.

The **Devices Plots** tab contains graphical hardware information. This includes Spellman/SuperN (a log of voltage and current values for users with a software controlled generator), CCDSCAM (a plot of CCD temperature for Eos, Atlas and Titan detectors, FIP60 (the same plot for Sapphire, Ruby and Onyx detectors), Cryojet (a plot of temperature and flow rates against frame number) and two further charts for the XrayChiller and CCDChiller.

This window is the point of access for all post data reduction data manipulation functions. This facility is labelled **Refinalization** and includes the application of numerical (face-indexed) absorption corrections, Laue setting changes, space group manipulation, data filtering and data truncation. **Inspection and Manipulation of Data** is described in detail in **Chapter 8**.

4.4.5 Launch 2nd instance of CrysAlis^{Pro}

This opens a second offline version of CrysAlis^{Pro} and transfers all current settings, such as experiment name, machine set up and unit cell information to the second programme, to allow for an independent, off-line processing of the data that is separate from any work done in the on-line window.

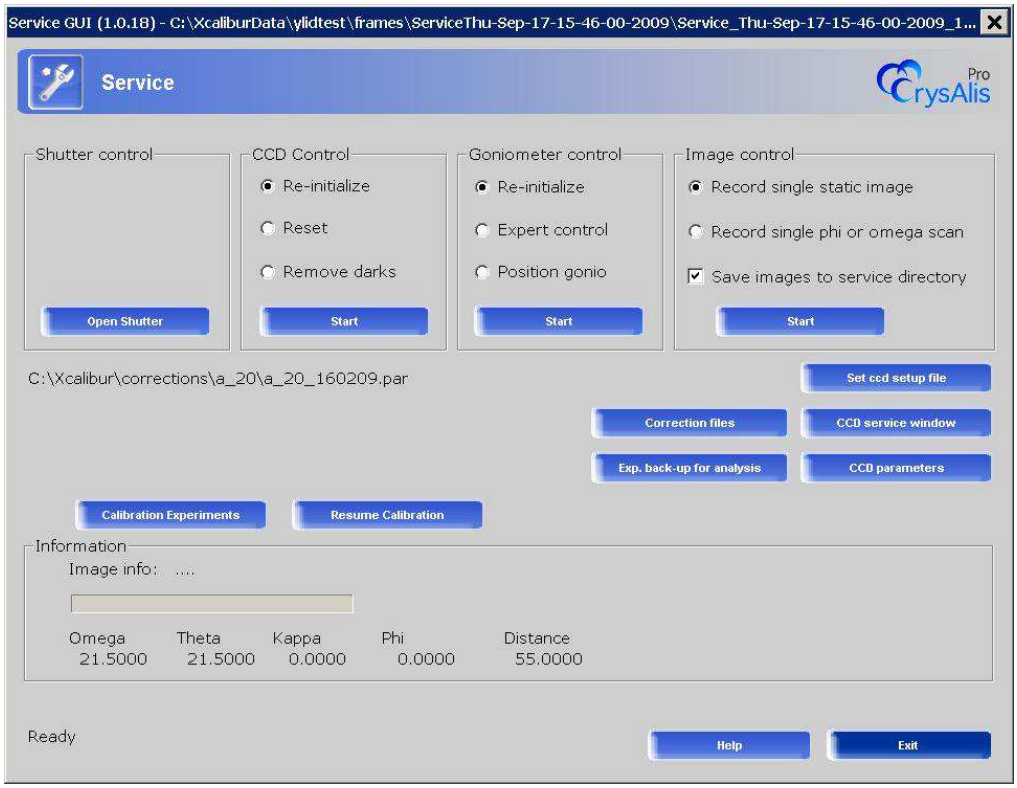


Figure 5 Service utility

4.4.6 Service

This power tool collects those service commands most commonly accessed by the user into one defined area. These commands include shutter open and close control, goniometer reinit(ialisation), CCD reinit(ialisation), Clearing dark image buffer (card clear Dark), direct goniometer control and home flag search (Mgcutil). It also provides for the definition of the machine parameter setup file. If the system interface is ever switched off, it will be necessary to reinitialise the goniometer using the service facility, by simply selecting **Gon reinit** and **Start**. Problems involving homing of the circles and detector require the **Mgcutil** or **Expert Control** facility. However, it is advisable to contact a service engineer or email XRDSupport@agilent.com before attempting manual goniometer control, as no collision protection is provided. CCD problems can often be fixed by selecting **Card reinit** or **Reset FIP** and **Start**.

More details on the service utility features may be found in **Chapter 7**.

WARNING

The mgcutil/Expert control window is the only place within CrysAlis[Pro] where there is no goniometer collision protection. Although warning messages will appear when driving the circles using mgcutil, the facility should only be used by experienced users or qualified Agilent employees.

4.4.7 Powder diffraction

The powder diffraction power tool performs different functions depending on the CrysAlis^{Pro} operating thread (**CCD/RED/USER**). When in **CCD** view, a powder experiment dialogue box opens. Here, the user can specify settings such as detector distance, exposure time, resolution and phi scan constraints to generate a run list for a powder diffraction experiment. To begin the experiment, click the 'Start with analysis' button at the bottom of the window. When the experiment is complete, a powder graph will be automatically generated from the correlated diffraction images.

If the powder diffraction button is clicked when in either **RED** or **USER** threads, a dialogue box appears asking if the user would like to extract the powder pattern from the current experiment. This function allows the user to generate a powder diffraction image, generated from the frames of a single crystal diffraction experiment. An image is built up as the frames are processed, and a powder graph is displayed upon completion.

Specialist commands that can be used to process specific areas of a powder diffraction image include **powder azim** and **powder radial**. The areas defined with the command will be greyed out on the diffraction image concerned. Full details of how to use these commands are available in the CrysAlis^{Pro} help utility under **Command Overview**.

Any output powder plots are stored in the experiment root directory as an experiment_powder.dat file. This is in text format and can be input into many specialist powder applications.

More details on powder diffraction features can be found in **Chapter 7**.

4.4.8 Multiple Temperature/Wavelength Experiments

This icon accesses a module where experiments can be set up combining multiple temperatures and both Mo and Cu wavelengths. The facility is very simple to use, and full details are given in **Chapter 6**.

4.4.9 Single images

When using the CCD thread, this utility allows the user to take still images, omega and phi scans as well as axial photographs. When in either RED or USER threads, only the **Axial photographs** option is available.

More details on the features available in single-image mode can be found in **Chapter 7**.

4.4.10 WinGX launcher

This icon sends a command to launch the crystallographic structure solution and refinement software WinGX, where installed. The current experiment name and folder defined in CrysAlis^{Pro} are automatically loaded. Assuming the data has been processed, the *.ins, *.hkl and *.cif_od (machine cif) files will be imported into a directory within the main data directory with the path \struct\olex2_samplename. The user can then solve the structure using a choice of solution programs within WinGX.

4.4.11 CCP4 launcher

When installed, this button launches the CCP4 suite of programs for protein crystallography.

4.4.12 Olex^2 launcher

This button launches the crystallographic structure solution and refinement software Olex^2, where installed. Assuming the data have been processed, the *.ins, *.hkl and *.cif_od files will be imported into a directory within the main data directory with the path \struct\olex2_samplename. These files are imported directly into the program and the structure can then be solved using the tools on the right-hand side of the screen.

The new Olex2 version 1.1 is fully supported by CrysAlisPro. The user can also install several versions in parallel. The version which is currently used is selected in the tools options panel 'Programs'

4.4.13 Crystals launcher

When installed, this button launches Crystals (structure solution and refinement software from David Watkin at the University of Oxford).

4.4.14 Jana launcher

When installed, this button launches Jana (a crystallographic program focused on solution, refinement and interpretation of difficult, especially modulated structures). Developed by Vaclav Petricek at the Institute of Physics in Prague, this software is the industry standard when working with incommensurate/modulated structure data sets.

4.4.15 Autochem launcher

Autochem is a CrysAlis^{Pro} plugin for automatic structure solution and refinement, and is a purchasable option. Autochem is designed to work concurrently during data collection, but it can also be activated post data collection by clicking on the power tool icon. Autochem data files are located in the autochem_experimentname folder within the Struct directory.

More details on using AutoChem are found in **Chapter 7**.

4.5. Diffraction / image display window

The majority of the CrysAlis^{Pro} GUI is taken over by the image display area which provides for the display of X-ray diffraction images. These can be displayed in a variety of sizes accessible through the zoom in / out functions.

4.6. Window header bar

The window header bar is used to display the current thread displayed in the main diffraction window.

There are 3 threads in CrysAlis^{Pro}:

1. **CCD thread** – used by the diffractometer. New diffraction images are displayed in this thread as they are collected and read out from the CCD detector. Whilst the diffractometer is collecting data, functionality in this thread is limited to adjusting the colour table, look up table, resolution rings, STOP and viewing the information cards.
2. **RED thread** – this thread can be used to view whole or partial data collections. The RED thread can be used to view data as the diffractometer is collecting, as long as the automatic data reduction is not

active. When automatic data reduction is active then the RED thread allows the user to visualise the data reduction in progress.

3. **USER thread** – this thread provides for the user to look and analyse images / data while both the CCD and RED threads are actively employed in data collection and reduction respectively. All functionality should be accessible in this thread.

4.7. Status area

- Shutter Open / Closed

  This icon simply reflects the current status of the X-ray shutter. Open (Red) with X-rays exposed or closed (Green) with no X-rays exposed.

- X-ray Mo or Cu button

  This button is only visible on the Gemini system. It automatically selects the source when it starts up, depending upon what default has been set up. To change sources just click on the button and press OK on the pop-up window. When the power switches from one generator to the other it will ramp to default figures of 20kV and 5 mA. The power must then be manually raised to the desired operational settings.

Figure 6 Status indicators

- CCD status

  This provides feedback of the current status of the CCD thread.

- RED status

  This provides feedback of the current status of the Red thread.

- Progress bar

  This shows the progress of the current process, for example, the percentage completeness of the pre-experiment or data collection.

4.8. Device control

4.8.1 Key to device control icons

	• CCD detector head controller
	• Cryodevice controller
	• X-ray generator controller
	• IO device control (SuperNova systems only)
	• Liquid nitrogen level indicator controller

Figure 7 Key to device control icons

4.8.2 CCD detector head controller

Clicking on this icon opens the software device controller for the CCD detector head. The display should show the running temperature of the CCD and the % power being used to maintain this temperature. The temperature should normally be about -40 to -45 degrees Celsius and the % power between 40 and 65%.

4.8.3 Cryodevice controller

Clicking on this icon opens the software device controller for whichever Cryodevice is installed with the system. A right-click of the mouse button on this icon allows the cryodevice to be changed, allowing control of Nitrojet, Cryojet, Cryostream (600 and 700), Cobra, Helijet and Hotjet devices.

The most commonly used device is the Cryojet. On the top of the display window the actual temperature at the Cryojet head is monitored (in degrees Kelvin) along with the Heater power output (in %) as well as Heater voltage (V). Below these are indicators about the current set values for the temperature (K), heater status (on/off) and the flow rates for the shield and sample flows (lt/min).

To set a new temperature, first click on the Start button (if the device is not connected yet), then click on the Set button on the right, which allows you to input a new set temperature, turn the heater on or off and set the desired sample and shield flow rates. In typical operation, 10 lt/min is used for both the sample and shield flow rates when cooling to a new temperature, once the setpoint is reached the flow rates are reduced to 6 lt/min. To warm up the Cryojet to room temperature flow rates of 3 lt/min are used. All operations normally have the heater switched on. The status line updates the state of communication between the software control panel and the Cryojet. The “Log file” and “Start Log” buttons give the user complete control over recording the temperature throughout experiments, however, the temperature is automatically logged from the beginning of data collection by default. The “COM Settings” button allows for changing the communication port settings manually, should there be a need.

4.8.4 X-ray generator controller

Clicking the X-ray generator controller icon allows the user access to the generator settings. On the top of the display, the actual Voltage (kV), Current (mA) and Power (kW) is monitored. Below these are indicators for the current set values for Voltage and Current and the appropriate filament current (A) and power limits (kW) for each particular system. The “Emergency X-ray Off” button quickly shuts the generator down and stops the production of X-rays in an emergency. (The hardware interface needs to be reinitialised (switched off and on) after such an event before setting the next Voltage and Current). The Start / Stop button initiates or ends communication between the software interface and the generator.

If not in communication already, click on START to connect to the generator. Use the “Set kV and mA X-ray” button to set new values. Typical settings for both Molybdenum and Copper fine-focus tubes (Xcalibur and Gemini systems) are 50 kV and 40 mA (although some users prefer 40kV and 40mA for Copper). For Microfocus systems (Mova and Nova), standard generator settings are 50kV and 0.8mA. The generator will reach the desired values in a stepwise and controlled manner. If you close the window before the ramp process is complete then only the values at closure will be reached.

In the bottom half of the X-ray window display there are Fault and Status indicators that provide feedback for the state of the generator. In normal operation only the Status indicators should be active. Use the Fault indicators as guides to investigating hardware issues should there be any trouble with the generator.

A log file for the generator output is automatically recorded from the beginning of the experiment and this can be viewed at any time by the Plot button.

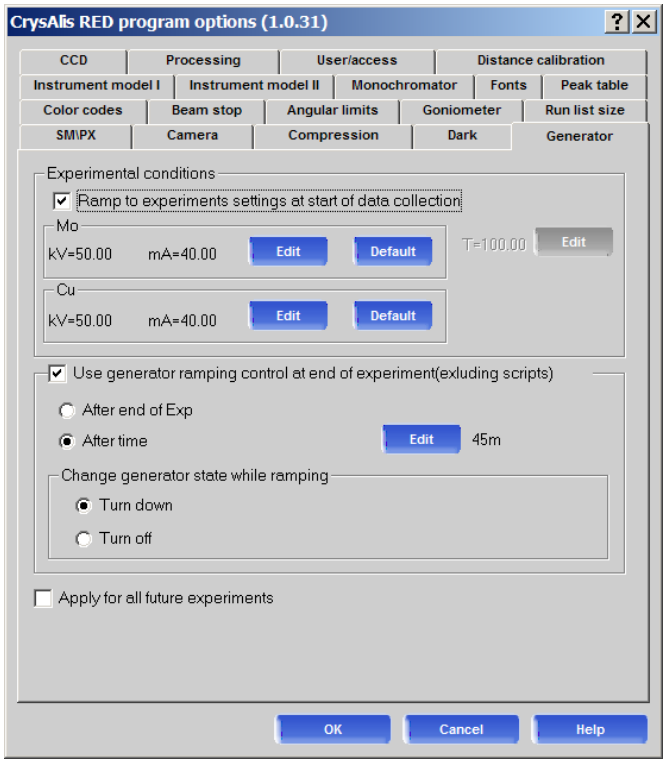


Figure 8 Generator ramping control

Clicking the right mouse button on the X-ray button gives access to **Generator options** (Figure 19). This menu controls automatic generator ramping before and after experiments. The generator can be turned down (or off) either immediately after an experiment or after a period of inactivity. If desired, the generator will also turn itself up to operational settings (user defined) when a new experiment begins. This facility is particularly recommended for Micro-focus systems, where keeping the tube at a standby power setting when not in use will prolong the lifetime of the tube.

4.8.5 IO device control

This IO device control button brings up a window with access to software control of the cabinet lights, and resetting of the emergency stop button.

4.8.6 Liquid nitrogen level controller

The liquid nitrogen level controller icon brings up a window panel indicating the level of liquid nitrogen in the tank (if there is an autofill connection). The level is expressed as a percentage, with 100% indicating that the tank is full.

4.8.7 Right Mouse Controls

Right mouse controls only work if the image control icon "i" (pixel/area information) is NOT selected. Using the right mouse button on the diffraction image displays a menu with various options:

- Pixel integration
  Select a peak by hovering over it, then right-click. Information such as integrated I/sigma and so on is displayed so that you can check that you have the correct exposure time
- Single frame peak hunting
  Select this to find all peaks in the frame and add them to the peak table
- Add peak
  To pick out individual peaks to add them to the peak table, put the cursor over the peak and right-click to add the peak
- Pixel value replacement
- Copy selection
- Remove background
  This clears the background noise from the diffraction image
- Semi-transparent windows on/off.
  This switches all semi-transparent windows between non-transparent and semi-transparent

5. Normal operation

In normal operation CrysAlis^{Pro} is designed to work in automatic mode. The automatic mode consists of the following three major components:

1. **Pre-experiment** – a short experiment to determine the X-ray diffraction quality of the crystal.
2. **Strategy** – determination of the best method of collecting the data under certain settings.
3. **Data collection**, data reduction and finalisation.

Before running a pre-experiment it is advisable to check the generator settings and the Cryojet control to ensure that the hardware is optimally configured for an experiment. To check these settings, click on the Cryo and X-ray buttons.

5.1. Crystal mounting and centring (F12)

To mount and centre a crystal, press the F12 key. This transfers goniometer control from the PC to either the manual control box inside the cabinet, or to the abs pilot module (in SuperNova systems). Centring of the crystal is easiest when the goniometer is in the **lower** position.

This same facility can be accessed by clicking the **Mount Sample** button in the pre-experiment window. A detailed procedure for crystal mounting is described below.

5.2. Pre-Experiment

1. To start the auto mode click on the **START/STOP** button in the top right corner of the GUI.

   A dialog will open giving the user a range of options.

2. Click on **START NEW** and a set-up window will open.

   By default an incremental experiment name such as Exp_1, Exp_2 *etc* will be assigned and a new data folder will be created in the root directory C:\Xcalibur data folder. The user can choose to change both the root directory where the data are collected and the experiment name by clicking on the 'Browse for root directory' button and typing the experiment name in the text box next to the label 'Name:'. Pressing **Return** at this point may start the experiment. It is not possible to overwrite existing experiments, however, the button 'Clear folder' allows the folder to be removed such that the experiment name can be re-used. In the browse dialog, usual windows operations such as rename, delete etc are accessible under the right mouse button.

   If you have any prior knowledge regarding the crystal it is a good idea to enter it in this window. The **chemical formula** for instance will be carried through the program and used in the absorption correction, *.ins file and summary report. The **chemical formula** will recognise brackets and common abbreviations such as Me, Et, Ph.

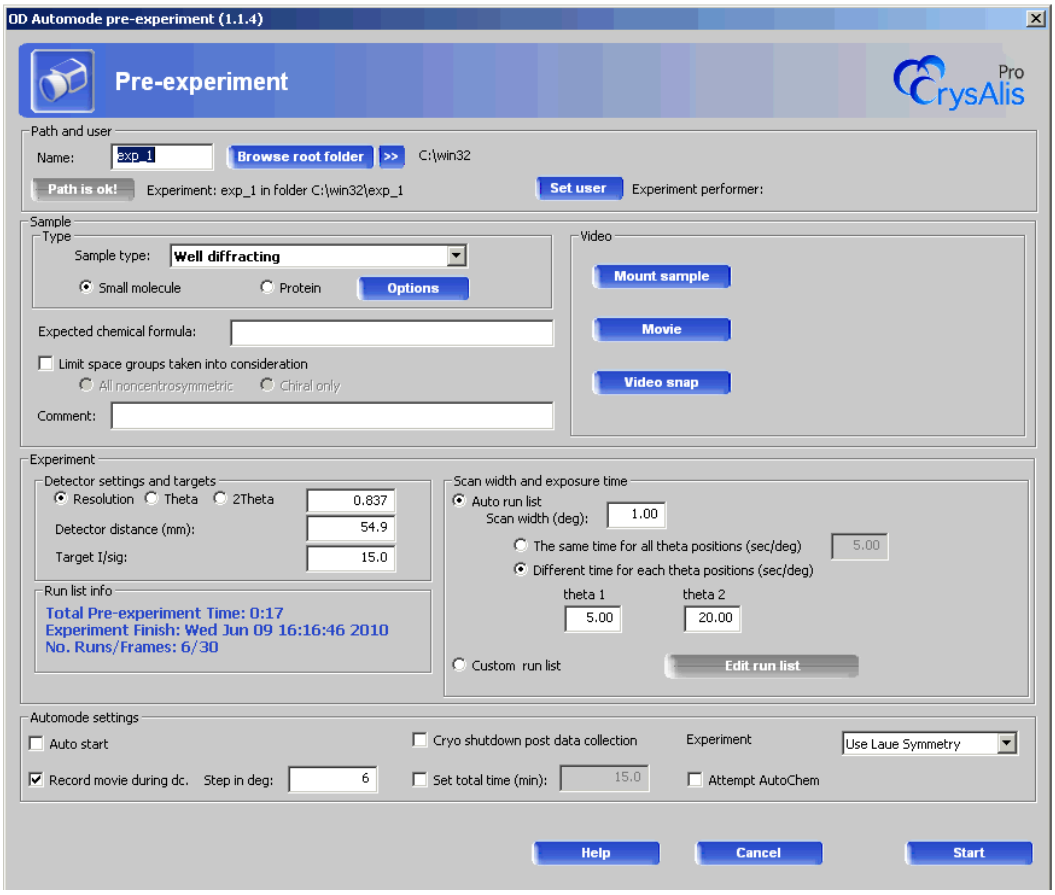


Figure 9 ***Pre-experiment setup dialog window***

3. Click on the **Mount Sample** button to mount a sample

> **WARNING**
> **Press 'STOP' on the remote control on the keyboard to stop movement of the equipment in an emergency. Mechanical movement of the goniometer and CCD detector may be performed using the remote control.**

- Pressing the mount sample button releases control from the computer to the remote control unit in the diffractometer cabinet.
- Press the **0** and **HOME** buttons on the remote control to drive the goniometer angles to the zero / home position.
- Mount the xyz goniometer head with attached crystal. Typically the crystal is glued on top of a glass fibre.
- Press **Lower** on the remote control. This will drive the goniometer to the correct orientation to allow optical alignment of the crystal.

NOTE

The settings lower and upper refer to the glass stick position on the video monitor

- Use the tool provided with the goniometer head to adjust the vertical height and horizontal position of the crystal, such that the crystal is in the centre of the video monitor screen.
- Press **180** on the remote control to rotate the crystal through 180 degrees. If the crystal's horizontal position has moved on rotation, adjust the position by approximately half the distance moved and in the opposite direction to the movement. Press **0** and repeat this procedure until rotation gives no movement of the crystal.
- Repeat the above process, rotating between **90** and **270** degrees.
- Press **Upper** on the remote control. The goniometer will now move to the upper position such that the goniometer head is located behind the collimator. If the vertical height of the crystal has changed, adjust as above and return to the lower position. Repeat until the vertical position is unchanged between the upper and lower positions.
- Press **Lower** and check alignment of the crystal on rotation between 0 and 180, 90 and 270 degrees.
- Press **0** and **Home** to return the goniometer to its zero position.
- Exit the alignment procedure by clicking close on the computer screen. This will return goniometer control to the computer and prevent use of the remote control.

4. Click on **Movie** or **Video Snap** if you want to record a movie of the crystal as it is rotated through 360°, or a picture of the crystal.
5. The information on whether the crystal is a **protein or small molecule**, the **crystal size**, **cell volume**, **diffraction limits** and if it is a **low temperature data collection** are used to adapt the pre-experiment which will be collected (you can see the changes being made in the experiment area in the middle of the dialog window). By defining the crystal sample as **Protein** or **Small molecule**, certain default parameters are employed throughout the pre-experiment, data collection and data reduction. For example, by choosing **Protein**, a **Smart background** calculation is employed in data reduction, whereas for **Small molecule** the default is for an **Average background** calculation. Protein data reduction also involves a default 2-cycle 3D data reduction.
6. The pre-experiment will typically carry out a 5-20 minute data collection in order to determine the crystal quality and to gather information required to design a suitable data collection strategy. For Mo this pre-experiment employs 3 orthogonal runs of 5 frames each. For Cu, where typically several theta settings are employed, there are more runs and more frames to collect.
7. The pre-experiment run list can be edited by hand using the **dc editruns** button. A preferred detector distance can also be chosen by clicking on the '**Detector = ?? mm**' button.
8. The **Scale exp. time** button allows the user to apply a scaling factor to the exposure time which is automatically chosen by the strategy software. The **Scale Exp. Time (theta shells)** button allows separate scale factors to be applied to pre-experiments where there is more than one theta setting. This is most important for Cu or high resolution Mo experiments. For example, if the exposure time is 5 seconds for all 6 runs of a Cu experiment collected to 0.84 Å, setting scale factors of 2 and 8 will give 10 and 40 second exposures for low and high angle theta positions, respectively.

9. The automode settings area of this window allows the user to put some restraints / constraints on the final data collection strategy. The **Auto Start** tick box (when selected) instructs the software to automatically continue with the full data collection at the end of the pre-experiment.
10. **Record Movie during dc** (if activated) will automatically record a movie of the crystal at the start of the data collection. This movie can be used later for face indexed absorption correction if required. The goniometer is driven to the lower position such that the phi axis and crystal are orthogonal to the video microscope. A series of jpeg images are then recorded at intervals (user defined between 1 and 6 degrees, 6 being the default) about phi, giving 360 degrees of photos around the crystal.
11. **Set max time** allows the user to put a maximum time constraint on the data collection. For instance, if 15 hrs is input, the data collection will be limited to finish within 15 hours. Note this does not take into account the additional time incurred by overflow remeasurements.
12. **Experiment type** – The default here is 'Use Laue symmetry' (where this means use the automatically determined strategy giving 100% completeness). The user can also select 'hemisphere', 'sphere' or 'quadrant'.
13. **Target resolution** – This is the maximum angular resolution (or 'd' value) to which the data will be collected.
14. **Set target I/sigma** – This is the target I/sigma required for the data being collected and impacts the estimated exposure times required for data collection.

    The I/sigma estimate is based on an analysis of the intensities of reflections recorded during the pre-experiment, and on theoretical predictions of the scattering power of the sample as a function of resolution. The purpose of including the theoretical data is to correct the overestimation of count times observed in those cases where intensities close to the resolution limit chosen for the pre-experiment are very weak or not observed at all. This can happen when the count time used for the pre-experiment is too short.

NOTE

It is always best to choose a reasonable pre-experiment exposure time, plus it is advisable to always check the exposure times generated for the main data collection before proceeding

15. If **Autochem** is installed, this can be activated for concurrent data collection and structure solution/refinement by ticking the box in the pre-experiment window.
16. When screening crystals, to avoid inputting pre-experiment settings multiple times, use the '**Recall previous Pre-experiment settings**' button in the bottom left of the window. Then either clear the folder, or change the experiment name and click **Start**.
17. Click on the **Start** button to continue.
    - The dialog will close, the pre-experiment data collection will begin and a pre-experiment information card / window will open on the right hand side of the GUI. Within this card / window are displayed the user defined settings from the previous dialog and the unit cell of the crystal being measured.
    - As soon as one frame has been measured the software will attempt to find the unit cell, crystal lattice and evaluate the average I/sigma spread with resolution (this last is used to determine the best exposure time for the data collection). Each of these will be displayed in the Pre-experiment information card and will be updated after every frame.

- At the end of the pre-experiment the strategy module will open and will display the preferred data collection strategy based on the unit cell, detector distance, Laue class, user constraints, resolution etc. If the Auto Start option was ticked on then this strategy window will automatically close and the data collection begin. Otherwise the window will remain open until user intervention.
- Clicking on the start full experiment in the pre-experiment information card / window at any time will accept the currently displayed unit cell and jump to the strategy module without finishing the pre-experiment.

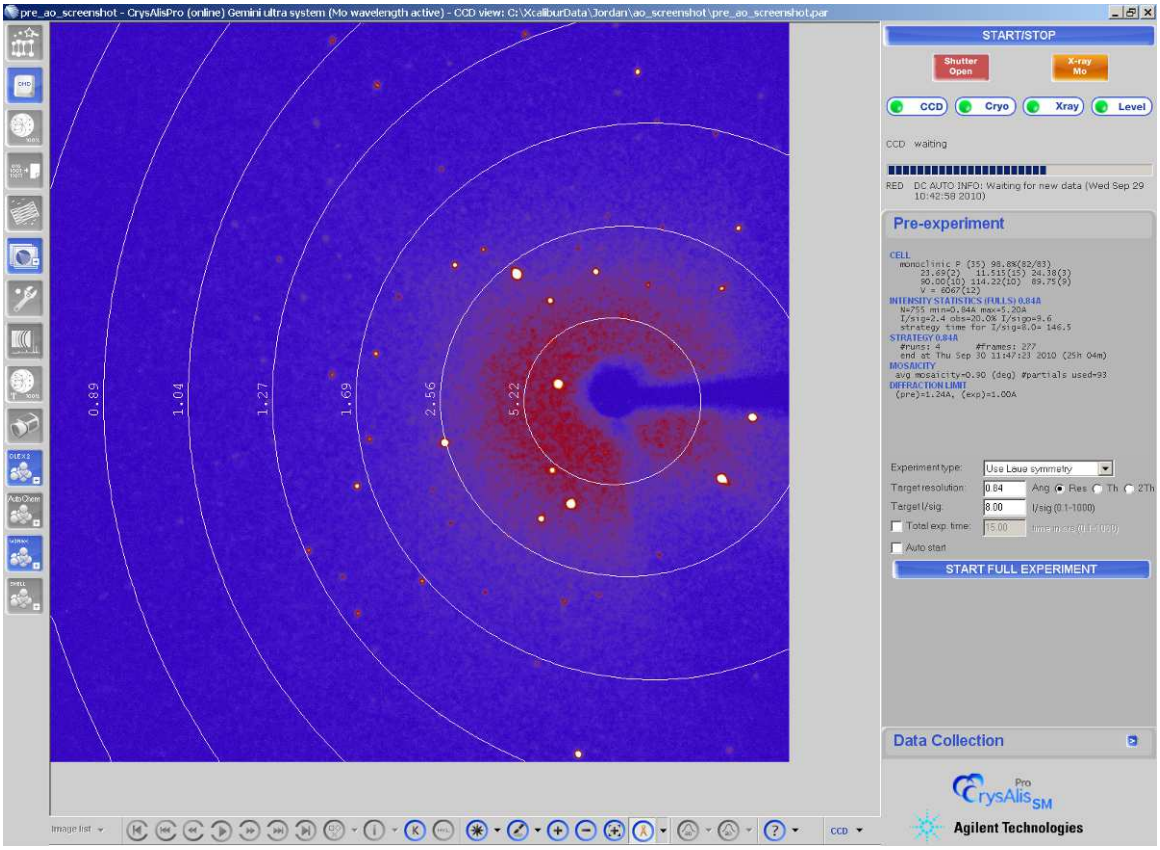


Figure 10 CrysAlis^{Pro} GUI, with active Pre-experiment information card

5.3. Protein Screening

The protein screening features can be accessed from the **START/STOP** menu.

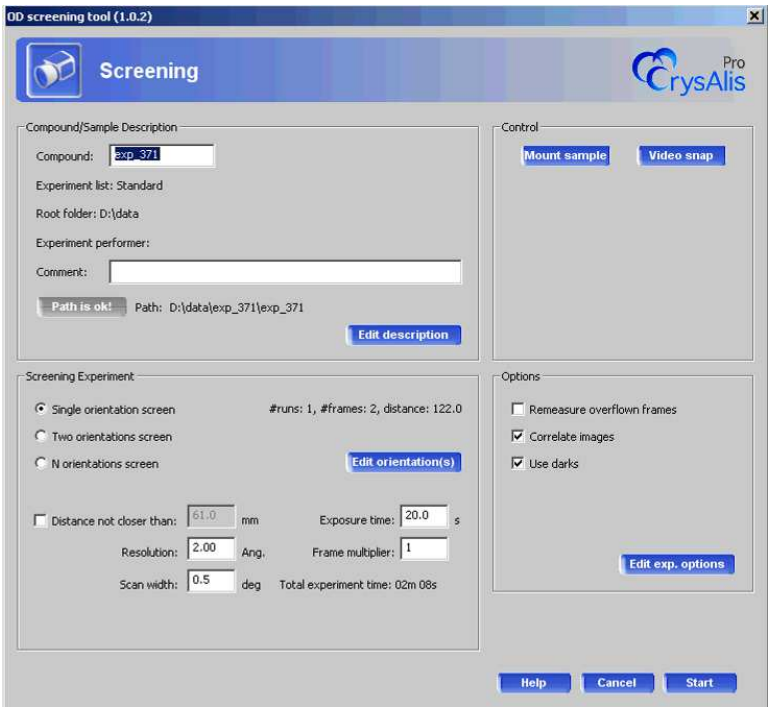


Figure 11 Protein screening dialog

Protein screening normally involves exposure times significantly longer than those for small molecule experiments. For this reason, the collection of dark frames at the beginning of a screening experiment can take a disproportionately long time compared to the frame of data itself. Unchecking the **Use darks** box will force the system to proceed immediately with the first frame of data. For the most accurate intensity data dark current subtraction should be always used. However, for qualitative work (sample screening, unit cell determination) the skipping of dark currents can significantly speed up your work. The screening results are summarized in the screening tab (figure 25), and there are hyperlink buttons on the screening tab to enable quick access to unit cell finding related functions.

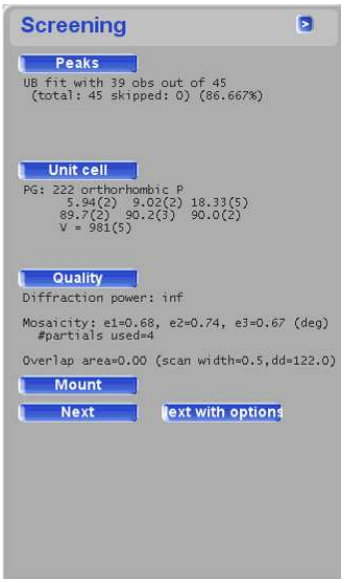


Figure 12 Screening information card

The **Peaks** button invokes the peak hunting dialog box. Here you can quickly tune the peak hunting parameters in case of difficult samples.

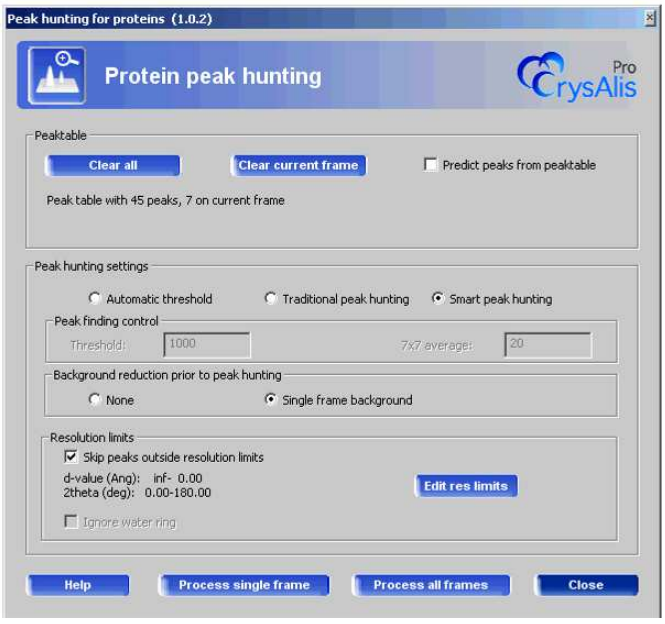


Figure 13 Protein peak hunting panel

The 2D drag tool gives an estimate of the expected d-values and possible lattice repeat distance (which could be one of the lattice constants.)

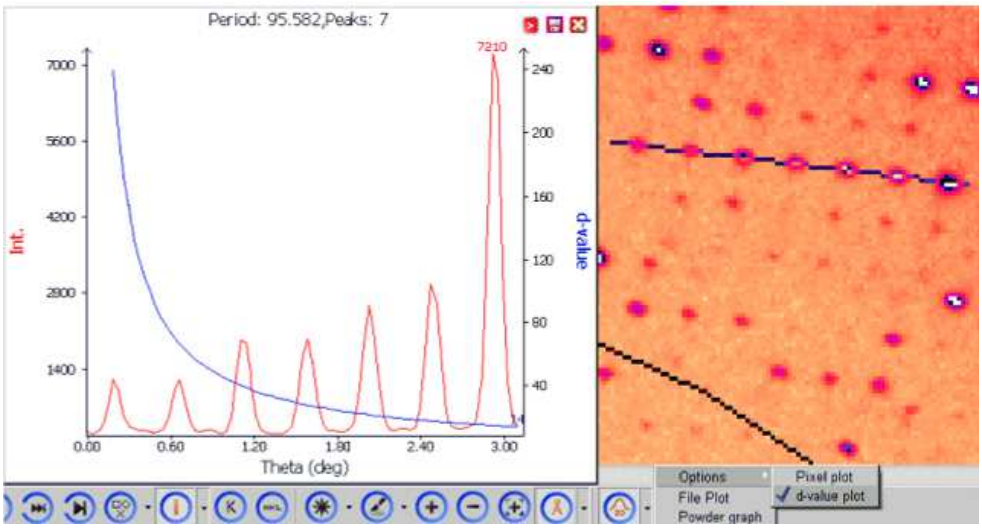


Figure 14 d-value tool

5.4. Strategy

The strategy module will automatically pick up the Laue class, detector distance, resolution target and exposure time from the pre-experiment and will determine an optimum data collection strategy with these and the unit cell in mind.

The default strategy is that required to obtain 100% complete data. However, a number of alternative modes are available. These include:

- 100% completeness
- Specified global redundancy with incompleteness allowed
- Specified global redundancy with 100% completeness
- Specified constrained experiment time with incompleteness allowed
- Specified constrained experiment time with 100% completeness
- Complete data for twins

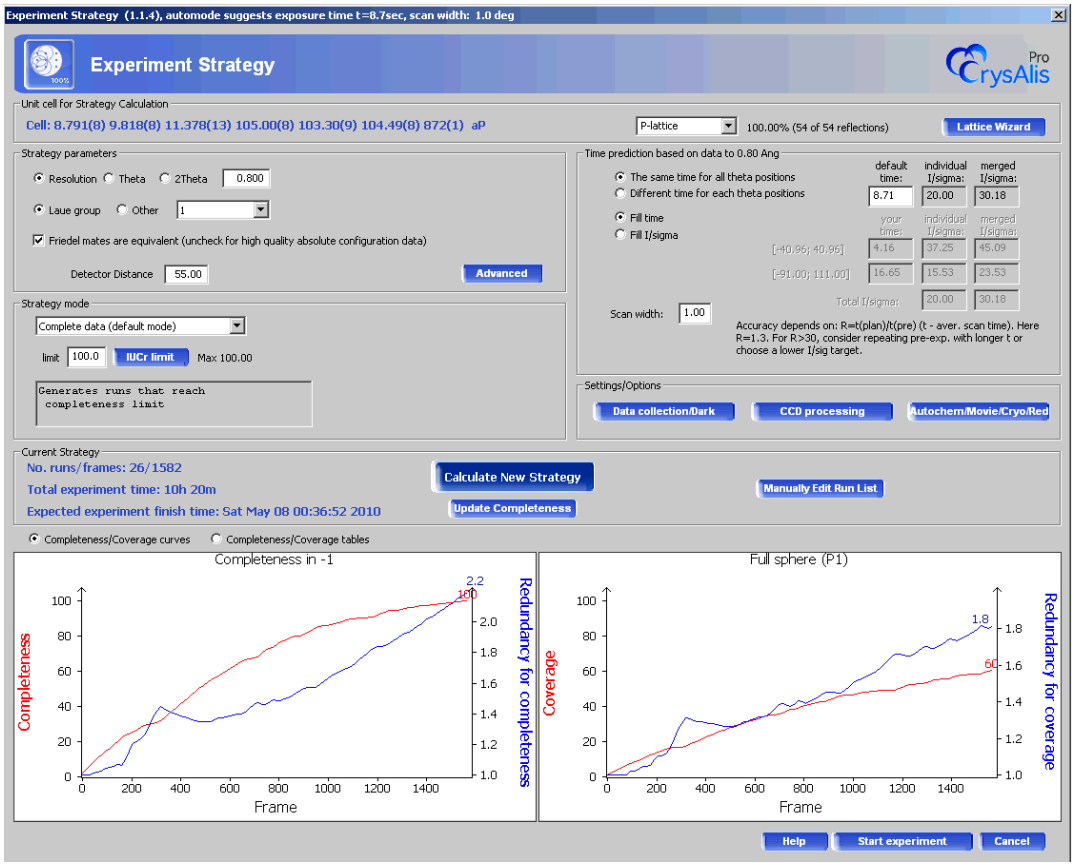


Figure 15 Strategy module

The strategy module displays two graphs. The one on the left depicts **completeness** and associated **redundancy** (related to the Laue class for the calculated strategy) and the one on the right illustrates the total **coverage** (% of the full sphere) and associated redundancy.

These provide a quick and easy graphical feedback. Alternatively, you can display the corresponding statistics tables by pressing the appropriate radio button just above the graphs. The chosen exposure time and corresponding scan width are displayed in the top bar of the window.

The strategy module provides access to a number of 'user parameters' which can be edited. These parameters include **resolution** (target max resolution of the data), **Laue** class, **Friedel mates are equivalent** and **detector distance**. If you change any of these parameters you must then click on the **Calculate New Strategy** button which will cause the strategy module to re-compute the best data collection module based on the updated parameters.

Friedel mates are equivalent should not be ticked if the sample is known to be or suspected of being chiral / non-centrosymmetric. This is particularly important in cases where absolute configuration is the main focus of the experiment.

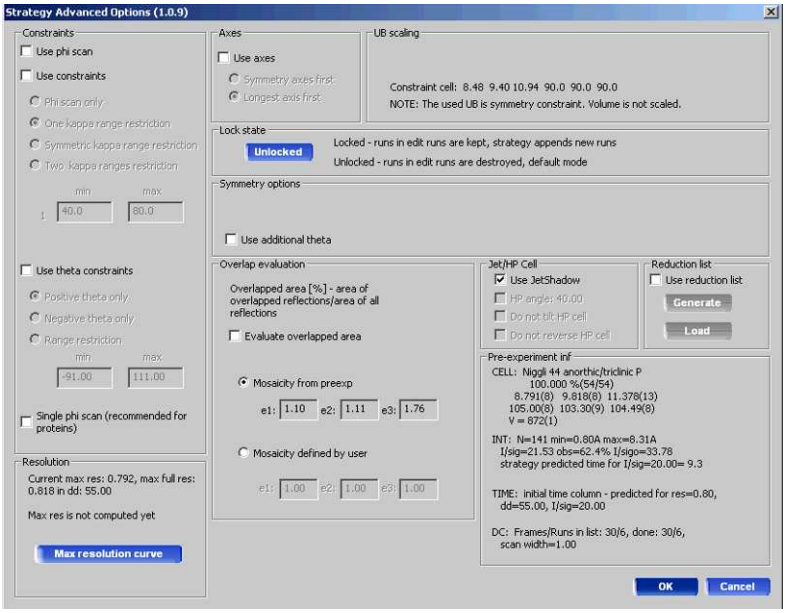


Figure 16 Strategy advanced options panel

The **Advanced** button opens an additional dialog window from which you can apply angular constraints on kappa/theta etc used during the determination of a data collection strategy. Cell centring can also be applied. The strategy can also be limited to only use omega or phi scans. The existing strategy can also be 'locked' and added to, and different exposure times can be chosen for each theta position (especially useful for high and low angle positions when using Cu radiation). After making changes click on OK followed by **Calculate New Strategy** in the main Strategy window. This will cause the strategy module to re-compute the best data collection based on the updated parameters.

The **Single phi scan** option found on the **Advanced** tab will produce a run with one phi scan in it. The strategy computation will adjust the scan starting point and range so that the highest possible completeness is reached in the shortest amount of time. It is intended primarily for proteins.

Also on the **Advanced** level you can specify a 'Reduction list' for the hkl generator. In this way you can collect data for a specific set of reflections provided you know their reflection indices. You can generate the list internally or load it from a user file.

If you have bypassed the pre-Experiment, but wish to set up crystal movie generation and AutoChem, you may do so by launching the Strategy module and pressing the 'Autochem/Movie/Cryo/Red' button located in the 'Settings/options' block of the Strategy panel.

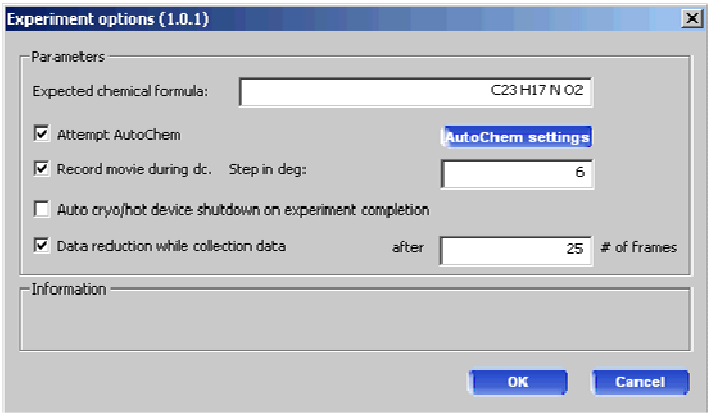


Figure 17 Strategy panel settings/options subdialog

If you are planning to process the image frames using an independent data processing software such as MOSFLM, you will need to ensure that the frames are always collected such that the scan rotation axis is perpendicular to the beam. The easiest method of ensuring this is either to select omega scans only or to fix the kappa angles. To do this, click on the **Advanced** button and tick on **use kappa limits**. Click the **one range** radio button. In the **min** box type the number 0 and in the **max** box type the number 0.000001.Click on **OK** which will close the window. Now click on **Calculate New Strategy** to calculate the updated data collection strategy.

If you want to change the automatically determined strategy click on the **Manually Edit Run List** button. This will open the runlist editor. From here you can alter the proposed data collection strategy as much as you like, selecting different exposure times, scan widths or completely different data collection ranges. When you have finished click on **OK** to exit this window and return to the strategy window. To see the effect your changes have had click on the **Update Completeness** button. The graphs and table of statistics should be updated.

When satisfied with your changes to the Strategy, click on Start Experiment to begin data collection.

5.4.1 Strategy calculation tips

The strategy module is a remarkably powerful tool and can be used in many different ways. Mastering the manipulation of data collection strategies is the best way to collect the best possible data. Here are a few tips related to common scenarios;

- Particular attention should be given to the exposure times used for data collection. Occasionally CrysAlis^{Pro} will overestimate the times required due to pre-experiment data being of low intensity (particularly at higher angles). The count times can be adjusted in terms of seconds or I/sig values. For Cu data sets collected to 0.84 Å (good data for IUCr publication), exposures for a high angle theta setting will generally need to be 3 or 4 times the value of those for the low theta setting. For high resolution Mo data, the same applies. The strategy module will use the same time for all theta positions as standard, so this must be changed by the user. All scaling of data to account for two or more differing exposure times is handled automatically.

- The strategy module will automatically adjust the scan width of the frames depending on the unit cell axis dimensions, mosaicity and overlap parameters found in the pre-experiment. The scan width can also be easily adjusted by the user. Generally speaking, for Mo radiation, cell axes greater than ~40 Å are often best collected with a scan width of less than 1°. Longer-wavelength Cu radiation can easily cope with much greater axis lengths. If twinning is suspected, reducing the scan width can significantly improve the chances of obtaining good quality data.
- When collecting data using an overnight time slot, it is often worth adding redundancy to a data set so that the whole amount of time is used. In order to add extra data, click **Complete redundant data** in the Strategy mode drop-down menu, choose a suitable redundancy multiplier (e.g. 5) and click **Calculate New Strategy**. Check the expected end time and recalculate as necessary. Having a higher redundancy means a better signal-to-noise ratio for the data and higher-quality determination of frame scale factors and empirical absorption correction.
- Some cryo-devices perform better than others and in humid conditions small amounts of ice can form on the metal pin below the crystal. This problem is enhanced when long exposure times are used and the goniometer head sits in or near the cold stream for a significant amount of time. When long exposure times are used (e.g. greater than 60 seconds), employing a kappa limit of -80 to +80 degrees will prevent any ice which may have formed getting close enough to the beam in order to be seen in diffraction images. Simply select **One kappa range restriction** in the **Advanced** strategy window and then calculate a new strategy. In most cases this will not affect the total data collection time, but will dramatically reduce the chances of ice diffraction.
- Various methods can be used in order to minimise the time taken for data collection. One of the most effective is to slightly reduce the complete data limit. Clicking on **IUCr limit** will reduce the figure from 100% to 98.5%. This may seem a small drop, but it has a dramatic effect. In most cases the time for data collection will be reduced by approximately 20-30%. This is because the extra reflections required for 100% completeness are in extreme places, requiring many extra frames to be collected. Adopting this tactic will also significantly reduce the redundancy figure, so although it is a good time-saving technique, it is better to collect 100% data if time permits.

5.5. Data collection

The data collection will start automatically using the strategy or user constraints. After 25 frames the data reduction will automatically start, followed by automatic frame scaling, automatic absorption correction, automatic space group determination and *.ins file creation. The progress of data reduction is shown in the data reduction information card. Data reduction will then wait 25 frames then repeat this process using all the data collected up to this point. This process will continue until the end of data collection.

Clicking on the external application power tools (WinGX, Crystals, Olex2) at any point during the data collection will copy the latest *.ins and *.hkl files into the Struct folder and open a project using these files, ready for structure solution.

5.6. Stop all processes

Working On-Line

1. Press the **START/STOP** button.

2. A dialog window appears with the option 'Stop All'. Press 'Stop ALL' and all processes will stop.

NOTE

Stopping all processes is also achieved by pressing the CTRL key on the bottom left of the keyboard!

- To pause an experiment, open the **Command Shell** and click **DC pause**. When ready to resume the experiment, click **Yes** on the dc pause dialogue box.

Working Off-Line

1. Press the **START/STOP** button.

2. A dialog window appears with the option 'Stop'. Press 'Stop' and all processes will stop.

If you do not want to stop all processes when the dialog box appears, then press the x icon in the top right corner to cancel.

5.7. Resume experiment/pre-experiment

In order to resume an incomplete experiment;

1. Click **START/STOP** and **Resume experiment/Pre-experiment**.

2. Click on the *.run file for the experiment, making sure to distinguish between the run files for the pre- and main experiments.

6. Non-standard operation

6.1. Start special collection

This utility is located under the menu accessed from the **START/STOP** button and is used for experiments generated without a pre-experiment. For example, if the user would like to use a previously generated run list, this can simply be imported and re-used. Generally speaking it is always best to conduct a pre-experiment with any new crystal. But in certain circumstances, especially when the same experiment is repeated under differing conditions, pre-experiments are not required.

To use this facility effectively, a cell must be defined, or a previously generated strategy imported. For the latter, this is achieved through the **Edit runs** utility by simply selecting **Import**. If the strategy has been used before the frames will be considered done, and so this must be reversed. Select **Refer to all runs**, then click **Invert done runs**. Return to the strategy window and choose **Completeness** to view your strategy.

To find a previously defined cell, click on the **Lattice Wizard** button and either load a peak table or parameter file from a previous experiment. The cell finding tools can then be used as normal. On returning to the strategy module, the chosen cell will be quoted at the bottom of the screen.

6.2. Append data collection

On completing an experiment, in certain cases it is useful to collect some extra data. Providing the crystal has not been removed from the diffractometer, frames can be added to the existing data set.

This facility is particularly useful in cases where a crystal is unexpectedly non-centrosymmetric, and the original strategy was calculated so that the Friedel pairs were considered to be equivalent. In order to publish such data, it may be necessary to collect more of the Friedel pairs.

1. On completing the experiment, go the **START/STOP** button and choose **Append data collection**.
2. In the resulting strategy window, select the **Advanced** button and make sure the **Lock State** says **Locked**. This means that the frames already collected are still part of the strategy but are considered done and locked. Any new strategy calculation will take these into account, and then add any necessary frames.
3. For the example described, keep everything else the same, but uncheck the **Friedel mates are equivalent** box.
4. Click **Find experiment** and a message will appear at the bottom of the window saying ‘x run(s) were considered locked for the finding of experiment! y run(s) were added!’.
5. Click **Start Experiment** to collect the extra frames.

6.3. Multi-Temperature/Wavelength experiments

This facility allows the user to queue up experiments for collection at a variety of temperatures and back-to-back Cu and Mo wavelengths. It is best used with a pre-defined strategy and so it is best to carry out a **pre-experiment** beforehand.

1. Conduct a pre-experiment as normal.

2. Define a strategy to collect as much data as is required for full evaluation at various temperatures. More data than normal may be needed in order to take account phase changes and other temperature effects.
3. Save the strategy as a *.run file by clicking **Export** in the **Edit runs** window.

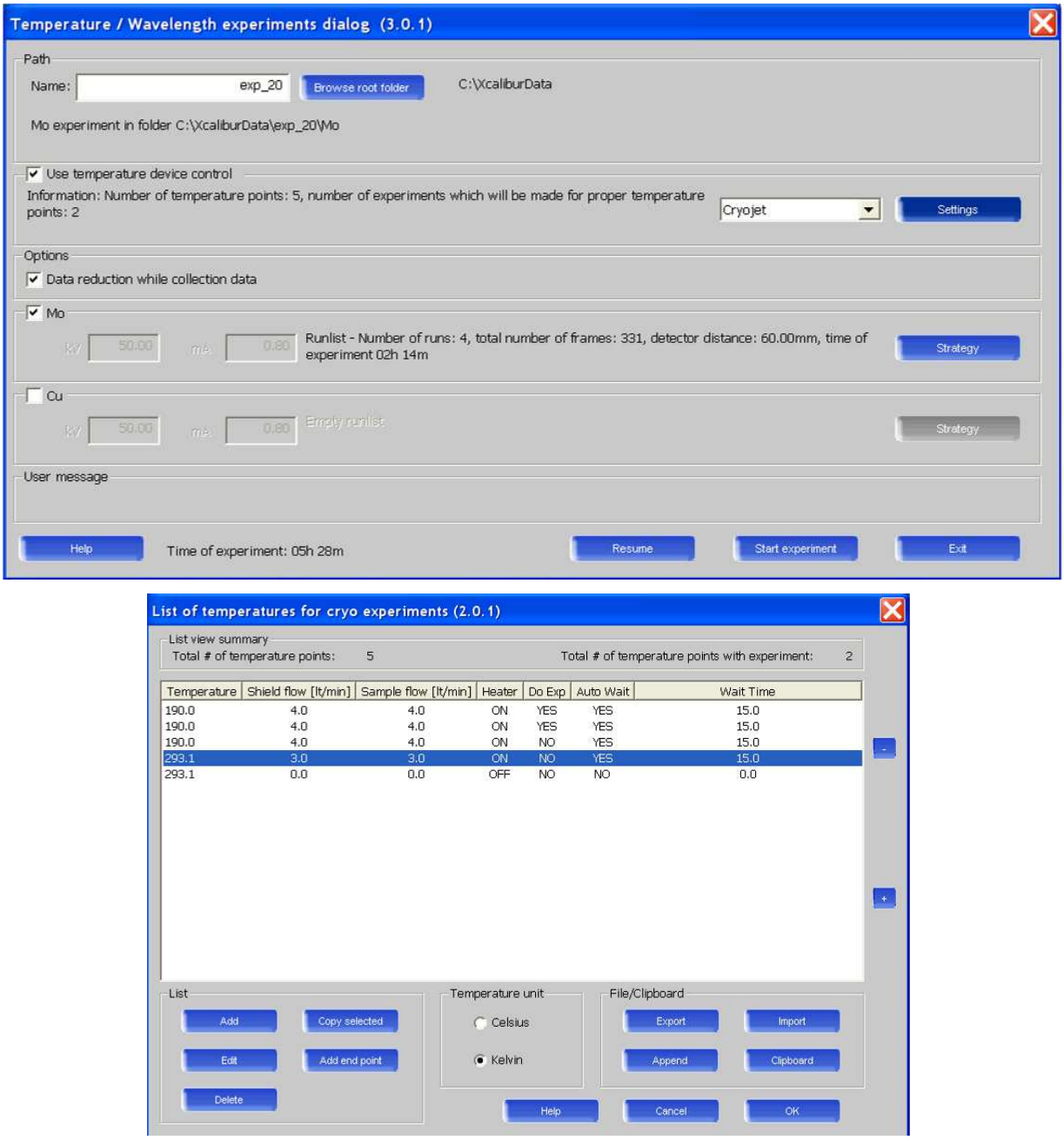


Figure 18 Multi-Temperature/Wavelength utility

4. Click on the **Multi-temperature/wavelength** icon.
5. For either Mo or Cu (or both), click on the **Strategy** button. Select **Edit runs** and then import the generated strategy.

6. Select the installed cryodevice and click **Settings**. By choosing start and end temperatures and a number of steps, a series of experiments will be run one after the other. For example, starting at 100K and ending at 300K with 5 steps will run experiments at 100K, 150K, 200K, 250K and 300K. These are stored in clearly labelled folders, with a temperature suffix.
7. The data is processed concurrently, but it is often useful to reprocess data in order to help correctly identify any changes in cell parameters over the given temperature range.

6.4. Data reduction – non concurrent

To reprocess data after data collection but using automatic settings, first open a second (offline) version of CrysAlis^{Pro}. Click on the **START/STOP** button and select **Load Experiment**. Select the experiment you are interested in from the experiment database list.

Click on the **START/STOP** button again and select **Full auto analysis(cell,red)** the program will find the unit cell including peak hunting etc and process the data, scale the data, find the space group and output the *.ins and *.hkl files.

If you have found the unit cell by hand using the **Lattice wizard**, click on **START/STOP** and select **Automatic data reduction**.

6.5. Data reduction with options

For problematic data sets it is often useful to process data with some manual influence over the settings used. The data reduction will be performed using the unit cell in the memory at the time and so the user must make sure they are happy with the cell before continuing.

1. If you would like to change the settings used by the profile fitting data reduction then click on **START/STOP** and select **Data reduction with options**.
2. The wizard allows the user to add lattice extinctions, omit frames from specific runs, and apply a variety of algorithm parameters and background calculations. If the data is weak or has a high/varying background, the data can be improved by incorporating the **Smart** background tool. A range of frames over which more localised background averages are applied must be chosen (odd numbers only).
3. When performing data reduction with manual settings, ensure that you click the button marked **Clear all data from previous runs**.
4. Sample wobbling in the beam can be accounted for by choosing options for either moderate or significant wobble.
5. The **edit special pars** window contains some specialist options for high pressure experiments, and enables the user to override the size of integration masks in cases where reflection overlap or mosaicity cause problems in the integration.
6. Outlier rejection is not important at this stage as this can be changed by Refinalizing later. However, non-centrosymmetric cases should have the Friedel mates box unchecked.
7. Space group determination can be done in either automatic or manual (interactive) mode by clicking the appropriate radio button on the last panel which appears. Using interactive mode can be very helpful in

ambiguous cases. For example, where an orthorhombic cell is found, but is transformed by the user to a monoclinic cell with the β angle very close to 90°, automatic space group selection will often choose an orthorhombic space group and transform the cell accordingly. Interactive space group selection can also be enabled from the Refinalization tool (See **Chapter 8**).

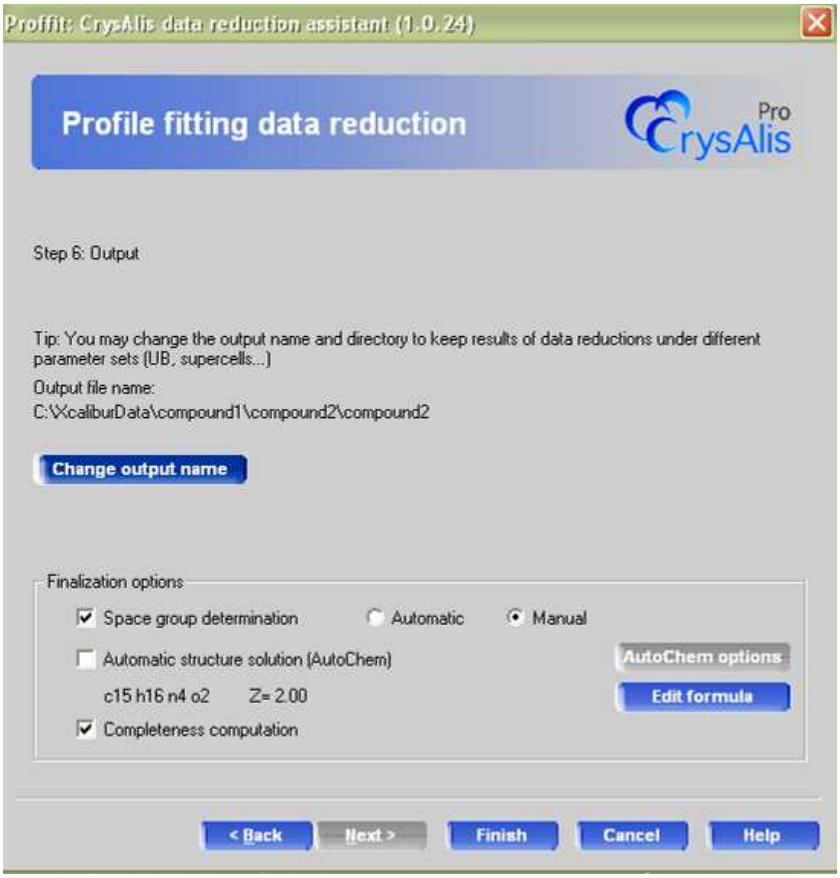


Figure 19 Enabling Manual (Interactive) Space Group Determination during data reduction

6.5.1 Tracking crystal movement

Occasionally a crystal will change orientation during a data set. This is usually related to glue which is not properly set, or sometimes needle-like crystals mounted on fibres or loops with insufficient points of contact. Crystal movement is easily observed by looking at reference frames from a data set (any change in the reference frames denotes a movement in the crystal or crystal decomposition). Also, if a crystal has moved the Ewald Explorer module will show two or more lattices which appear to be in different orientations (also a sign of non-merohedral twinning). However, such data sets can be recovered using a tool which looks for changes in orientation matrix from one run to the next.

1. Peak hunt on the first run only (or possibly the pre-experiment only) and use the cell finding tools to find the initial cell and orientation matrix.
2. Peak hunt over the full data set, but do not attempt to index all the reflections using the initial cell.
3. Choose **Data reduction with options** in the **START/STOP** menu.

4. At step 3, tick all three boxes, the most important of which is **Follow sudden (discontinuous) changes in sample orientation**. Set the **Orientation search range** and **Steps per degree** to suitable figures (higher values mean a longer processing time but a greater chance of success).

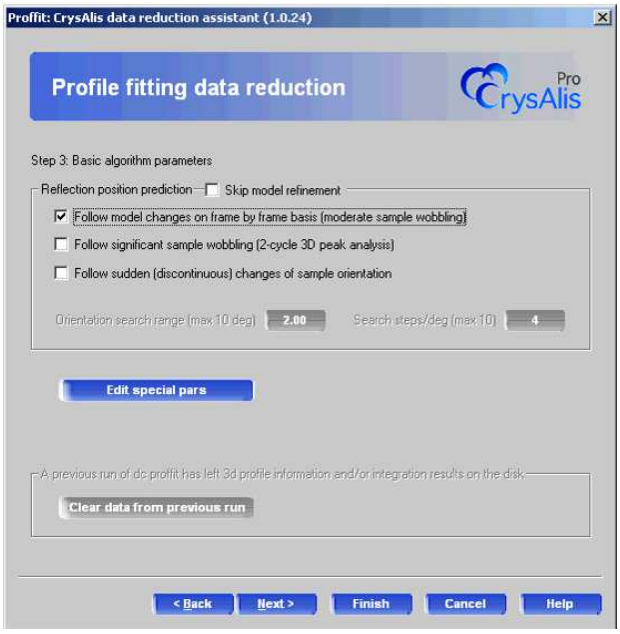


Figure 20 Follow-model options during data reduction

5. If successful, the Ewald Explorer will show one single and good quality lattice. There should also be a remarkable improvement in R_{int} statistics.

7. Power Tools

7.1. Lattice Wizard

The lattice wizard contains all the tools necessary for finding the unit cell and Laue group. The tools within the lattice wizard group are discussed in turn below:

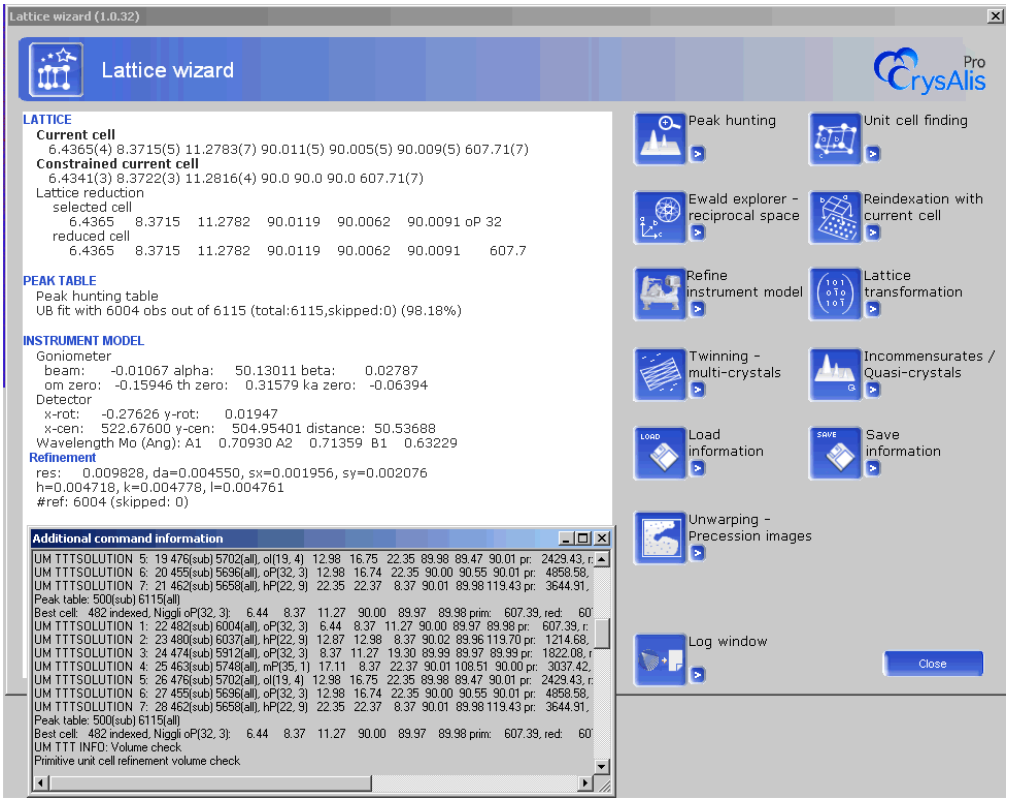


Figure 21 **Lattice wizard panel including log window**

7.1.1 Peak hunting

Click on the peak hunting icon and the computer will go through all the peaks and extract all those above a threshold level. When this has finished it will return to the main lattice wizard screen. In the window there will be a report on how many peaks have been extracted. To access user settings such as thresholding and smart peak hunting, press the arrow button to the right of the main peak hunting icon. The menu options are:

- Peak hunting with user settings
- Auto analyse unit cell

Under the peak hunting with user settings menu, the **Smart** peak hunting option is particularly useful. This will generally find 20-30% more peaks, but without picking up too much background noise.

7.1.2 Unit cell finding

Use the unit cell finding icon after you have finished peak hunting. Press the unit cell finding icon and the computer will automatically determine the unit cell. Information will appear in the top left of the window. More options are accessible by pressing the arrow button to the right of the main unit cell finding icon. These include:

- Unit cell finding with cell options
- Solution selection unit cell finding
- Search for known unit cell
- Set orientation matrix by hand
- Search for smaller unit cell volume

Unit cell finding with options allows the user to set limits on the minimum and maximum axis lengths to search for. This is also the utility used for twin lattice finding (see **Chapter 9**).

Solution selection unit cell finding gives a list of cells considered by automatic unit cell finding. If looking for a specific cell which is not output by automatic cell finding, it can often be found in this list.

Search for known unit cell allows the user to input cell dimensions and search using a specified number of reflections. The **Stop if % >** figure should be set to no greater than 30-50% for the best chance of success.

7.1.3 Reciprocal space visualisation – using Ewald Explorer

The peak table can be displayed by selecting the appropriate option which appears after pressing the small button next to the Ewald Explorer tool. Optionally, the peak locations can be cast in terms of diffractometer setting angles, Cartesian coordinates, or coordinates in the detector frame of reference.

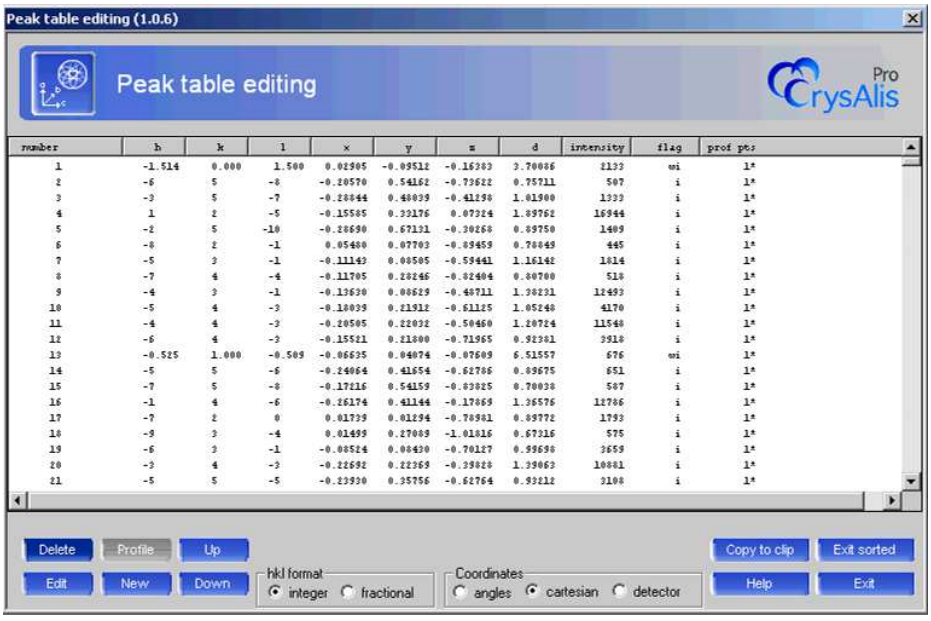


Figure 22 Peak table panel

Alternatively, the current contents of the peak table can be visualised using the Ewald Explorer reciprocal space visualiser. Ewald explorer allows the user to rotate reciprocal space, zoom in and out, drag-index and select

reflections for peak indexing. The user can also adjust intensity thresholds, lattice extinctions and d-spacing thresholds to select peaks. Information on using the Ewald Explorer to find multiple lattices in twinned crystals can be found in **Chapter 9**.

7.1.3.1. Drag indexing a unit cell

1. Click the Ewald explorer button.
2. The Ewald explorer allows you to scan the reciprocal space of your data collection for lattice planes. The image can be rotated by holding down the left mouse button whilst the pointer is positioned over the image, and rotating it.
3. Try to find a plane by rotation about x,y,z. A lattice overlay can be shown by ticking the **Lattice** tick box.
4. Click the radio button **Drag**, then the radio button **DragIndex**. This will allow you to specify three non-collinear vectors to define a UB matrix and thereby a unit cell.
5. Click on the button **b*-order** radio button and then left mouse click on the lattice and drag out a vector. The corresponding d-value is shown below the **b*-order** button. Count the diffraction orders you covered and enter the value by clicking on the button **order b***.
6. Repeat the previous bullet point for **a*-order**, rotate and repeat for the **c*-order**.
7. Click on the button **UM S** to set the UB matrix based on the DragIndex vectors, and exit the Ewald explorer using the **OK** button and select **Yes** if you are happy with your results.
8. Index the reflections by pressing the button **index with known cell** and repeat if necessary. A percentage figure of merit for the cell is displayed in the lattice wizard window.
9. Sometimes a negative and non-standard cell will result, so we apply a lattice reduction by using the arrow button under the lattice transformation window.

7.1.3.2. Drag-Selection of Reflections and Marking them as Skipped

1. Make sure that the **Select** radio box and the **Drag** radio button are selected. Depress the left mouse button, drag out a wire box around the reflections to be skipped, right mouse click and select **Mark selection skip**. These reflections will then turn red in colour indicating they are skipped. Any skipped reflection will not be used when unit cell finding.
2. Click on the ''used'' radio box. Only those white reflections to be 'used' in unit cell indexation (and not the red skipped reflections or wrong *ie.* Unindexed reflections) are now shown.

7.1.3.3. Intensity Selection of Reflections and Marking them as Skipped

1. Select **Skip filter** from the top menu and select **Intensity**.
2. A new window entitled 'Skip filter intensity' should appear. By clicking on the Max left and right arrows, decrease or increase the intensity threshold. As you increase the threshold using the right arrow you should see reflections disappearing in the main Ewald Explorer window. The Min left and right arrows also allow the minimum threshold intensity to be adjusted. Adjust the Max and Min settings to remove the unwanted reflections from view, ensuring that the 'Use intensity filter' box is ticked. Click on **OK**.

3. From the menu bar select the **Flags** menu and then **Mark invisible skip**. A window will appear asking you 'Do you want to mark all invisible peaks skip?' Click on **OK**. The 'invisible' reflections which have been selected using the intensity filter have now been skipped.
4. Clicking on the Show radio button 'skip' will not show these peaks since the intensity filter is still in use. To view these peaks go to the menu bar at the top of the Ewald explorer window and from the **Skip filter** menu select **Intensity**. The Skip filter intensity window will appear. Click on the **Use intensity filter** radio box until the tick disappears and the box is empty.
5. Click on **OK**. Select the Show **skip** button from the bottom left corner of the Ewald Explorer window. All the skipped peaks, including the intensity skipped peaks should now be visible as red dots.

7.1.4 Indexation with known unit cell

It is possible to re-index the peak table using the currently loaded unit cell. This applies a round of least squares, and shows the results in the lattice window. More options are available using the arrow button to the right of the icon. These include:

- Indexation with known unit cell and criterion
- Non-integer indexation
- Twin indexation

The first of these options allows the tolerance for indexation to be changed (set to within 0.125 of an integer value as default).

7.1.5 Refine instrument model

This facility enables the user to manually calibrate the diffractometer's instrument model. However, this tool should only be operated by specifically trained personnel.

CAUTION
Manual calibration of the instrument should only be carried out by an expert. The calibration of the diffractometer may be lost if an invalid model exists

7.1.6 Lattice transformation

Click on the main button to open the lattice reduction window. This allows you to select the particular cell settings and Bravais lattice from a list of possibilities. The arrow button to the right of the main button gives the **lattice transformation with user matrix** option, so that a matrix to apply a transformation to the unit cell can be entered.

7.1.7 Twinning - multi-crystals

A twin is formed when two or more crystals of the same material intergrow. The unit cells of the two (or more) components are related to one another by symmetry. The lattices may completely coincide in all three dimensions (merohedral twinning) or in fewer than three dimensions (non-merohedral twinning).

A twin may be suspected for a number of reasons:

- Re-entrant faces on the sample

- Non-uniform polarisation of light when examined by microscope
- Peaks which appear split in the diffraction pattern
- Difficulty with indexing, or a low percentage of indexed peaks
- Difficulty solving and refining the structure to a reasonable R_1 value.

If a twin is suspected before data collection, it is often beneficial to move the detector further back than you would for a standard data collection. This will help to separate the spots from each twin component.

Consider using slightly longer exposure times and collecting higher redundancy than normal to increase the number of data points.

Twin data decomposition is discussed in detail in **Chapter 9**.

7.1.8 Incommensurates / Quasi-crystals

An incommensurate or modulated structure derives from data where there is periodic distortion of the atomic positions (modulation) and/or of the occupation probability of atoms (density modulation). This gives rise to satellite reflections present in addition to the main lattice. Satellite reflections are often of very weak intensity and so are not always clearly observed. CrysAlis^{Pro}'s utilities for incommensurate data processing are described in detail in **Chapter 10**.

7.1.9 Precession photo reconstruction

This power tool allows the user to reconstruct precession photographs. This is achieved via a wizard that guides you through the process and can utilise either the complete data set or specific runs and frames.

1. The guided wizard is started by clicking on the icon **Unwarping - Precession images** in the lattice wizard.
2. The current orientation matrix is loaded. Click on the **Next** button to proceed to the next step.
3. If required adjust the number of runs / frames used in the unwarp reconstruction by clicking on the relevant run in the main text box. The whole line occupied by that run should be highlighted in blue. Now click on **Edit start num of selected run** or **Edit end num of selected run**. Enter the desired parameters. Setting the start number of a selected run to zero will prevent that run being used in the reconstruction. Click on **Next**.

NOTE

The greater the number of runs / data employed the longer the reconstruction will take

4. Click on the **Browse for output dir** button and create a new folder where the reconstructed images will be stored.
5. To generate the three most common layers (hk0, h0l and 0kl), click on **Generate layers**. Changing the order to 1 will also generate the -1 and +1 layers, giving 9 in total. Set the resolution and click **OK**.
6. To generate a specific layer, click the **New layer** button. Define the required layer for reconstruction using the 3 vectors. Select 2d Laue symmetry averaging if required. Use 2d Laue symmetry averaging when the symmetry of the crystal is KNOWN. This will enable faster and more complete layer reconstruction. Only the data required to define the unique part of the layer need be present in the data collection. Click on **OK**.

To set up the layer to reconstruct, define the plane using the L1 and L2 boxes. L1 1 0 0 defines the h direction and L2 0 1 0 defines the k direction, so both together define the hk0 layer. Define a name for the image in the Output name box, and then click **OK**. It is possible to define several layers to be generated as part of the same process.

7. If required click on the tick box marked **background subtraction**. Click on **Next**.
8. Click on **Finish**.
9. The defined layer or layers will now be reconstructed. A process box appears and the reconstruction can be seen in the image area when you press the **layer** radio button. The process box disappears when the process is complete.

Images can be opened for viewing in the image list menu on the image control icon bar. A reciprocal lattice grid can be overlaid on procession images by clicking on **Unwarp reciprocal lattice grid** in the drop-down menu next to the **CCD/RED/USER** button in the image control icon bar.

7.2. Service

The service utility window also provides access to the diffractometer calibration experiment tool. This facility should not be needed other than during installation and service visits, but if persistent problems are encountered, the user may be advised to recalibrate the instrument.

If required, an automated procedure is normally used, with data collected at two different detector distances (near and far) on a test crystal (an ylid test crystal is provided with the system). With Gemini and dual wavelength SuperNova systems, the procedure is carried out for both molybdenum and copper sources. In order to save time, calibration experiments are often run without frame correlation or overflow re-measurements, both of which can be switched on/off in the **Calibration Experiments** window. The experiment can be further optimised by changing the resolution and exposure times. The calibration can be monitored in the **Command Shell** and an output message is generated upon finishing which details the instrument parameters, test crystal unit cell dimensions and the percentage fit for indexed reflections.

From the service power tool we can access the automatic instrument calibration tool. Generally calibrations are run for two purposes:

- instrument model calibration
- instrument model calibration with additional flood calibration

Often the performance of the standard sample is a characteristic fingerprint of the machine performance. But the R1 statistic is really a function of the data completeness. To make the results comparable, an option has been added that will append to the calibration frames normal data-collection frames to produce a complete data set.

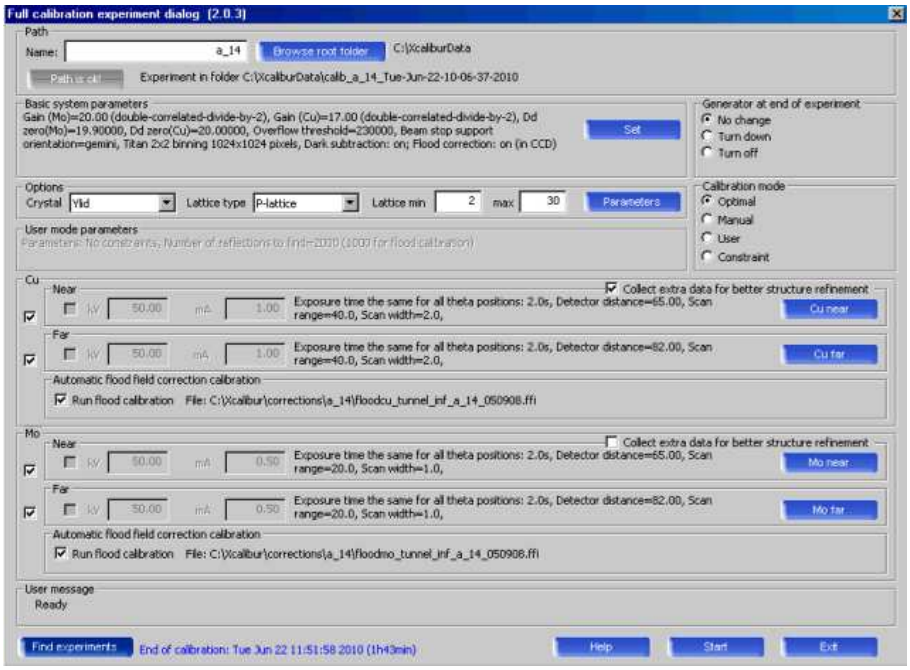


Figure 23 Setup Calibration Experiments

There is also a 'Make zip' tool. Users can zip up complete experiments into a single file for easier archiving or to send to the software team for problem resolution.

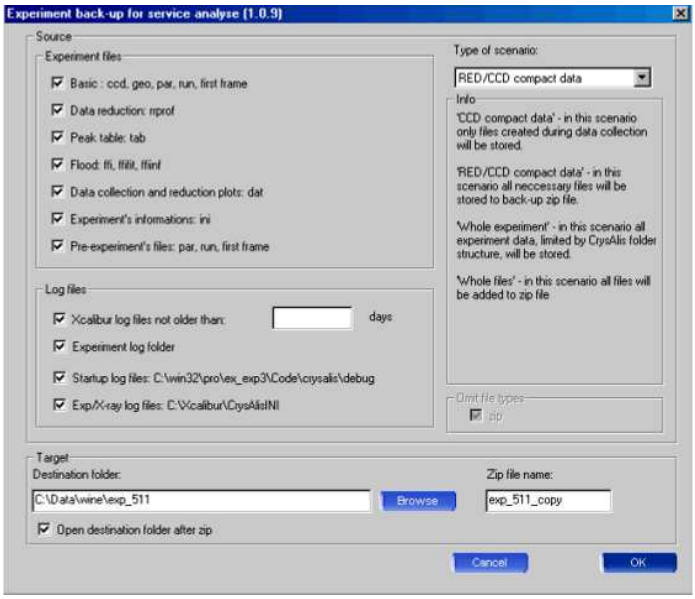


Figure 24 Experiment Archiving Facility

7.3. Powder Diffraction

When in CCD mode in CrysAlis[Pro] the 'Powder power tool' allows you to design powder experiments:

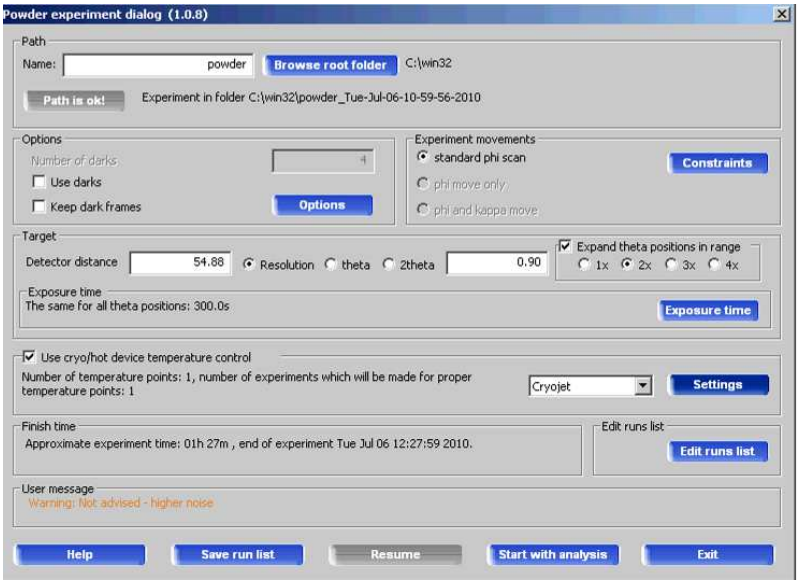


Figure 25 Powder power tool dialog

The user selects a detector distance and target resolution and the power tool sets up the experiment. Then the **Exposure time** control panel is opened so that the exposure time may be set:

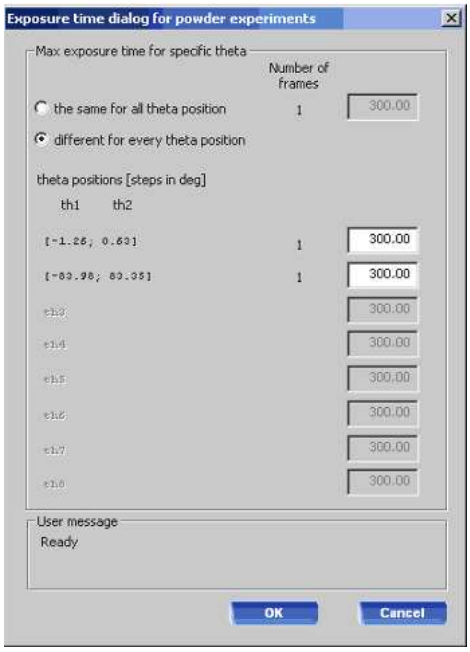


Figure 26 Powder power tool exposure time subdialog

For qualitative powder patterns the dark current is not essential. You can uncheck the 'Use darks' check mark under 'Options' to get faster powder experiments.

For calibration powder experiments you can use the 'Expand theta positions in range' check mark, which will insert into the run list more overlapping theta positions to see the same ring under different conditions.

7.4. Single Images

Clicking on the single images icon displays a box with options to:

- Record a still image
- Make a phi scan
- Make an omega scan
- Make a rotation photo
- Axial photographs
- Screening data collection

Only the axial photographs option is clickable when working off-line. It allows you to calculate the position at which you might take a photograph.

Click on an option to select it, and then click on the start button.

7.5. AutoChem

Olex2 version 1.1 is fully supported by CrysAlis^{Pro}. The user can also install several versions in parallel. The version which is currently used is selected in the tools options panel **Programs**, as shown in Figure 28.

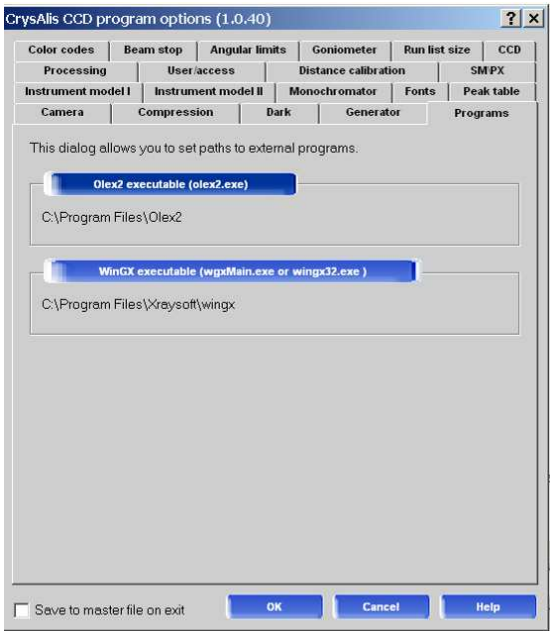


Figure 27 Programs tab of CCD setup options panel

Provided that you have installed AutoChem, you will have access to the AutoChem control panel.
We support at the moment the following solution methods: ShelXS, smtbx charge-flipping and ShelXD. As refinement methods we support ShelXL and smtbx-refine. Within Olex2 you can also install SuperFlip and SIR as a solution method.

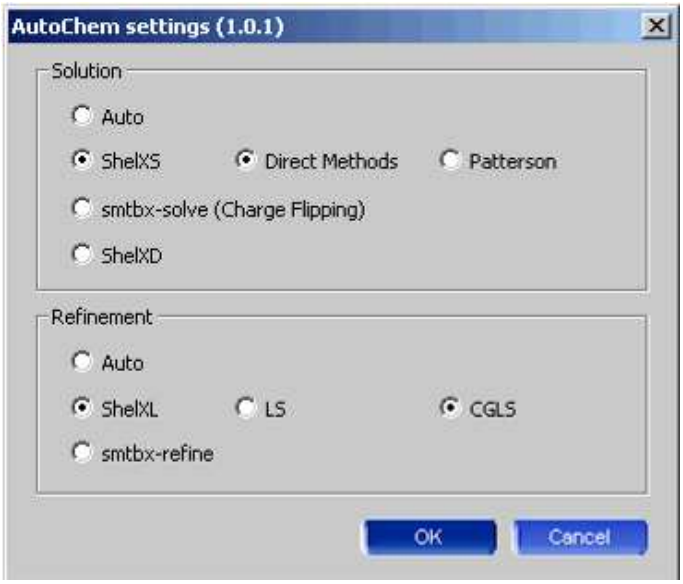


Figure 28 AutoChem settings panel

Both AutoChem and Olex2 now share the same structure folder. This makes it easier to review an AutoChem structure after concurrent data reduction. As a result of this we now have to halt the Autochem operation in concurrent data red while Olex2 is open. For this reason you might observe the following states on the AutoChem tab:

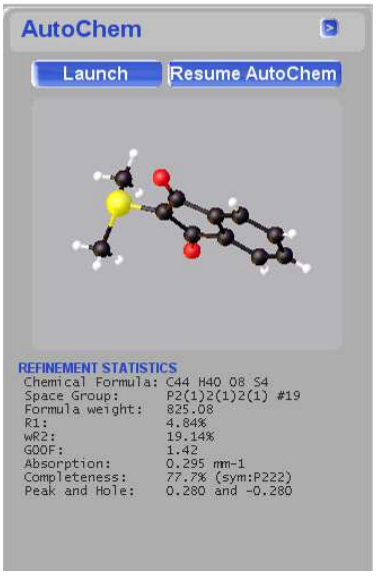


Figure 29 AutoChem information card

8. Inspection and Manipulation of Data

On completion of an experiment, the data reduction tab within the main CrysAlis^{Pro} window displays various output information. However, for more detailed results the **Inspect data reduction results and refinalize with user corrections** (4th power tool button) utility contains a wealth of information about the completed experiment.

The **Data reduction File contents** area contains a summary of cell dimensions and orientation matrix used for integration along with a run by run listing of scan type, angle ranges, exposure time, detector distance and the number of reflections collected in each run.

The **Data reduction output** tab contains the full log of the automatic data reduction process. Starting with the generator settings and data collection temperature, information is provided on any re-measured frames, profile fitting parameters, outlier rejection, scaling, space group determination and final merging statistics, allowing for a thorough inspection of all aspects of the data reduction process.

The **Data collection output** tab gives information obtained during the experiment, specifically relating to remeasured frames and overflows. Any problems encountered during the experiment will also be displayed in this log.

The **Red Graphs** window area displays a run-by-run visual summary of the scaling coefficients (absscale), a frame-by-frame plot of R_{int} and a number of comparisons regarding the coverage and or completeness of the data.

The **Devices Plots** tab contains graphical hardware information. This includes Spellman/SuperN (a log of voltage and current values for users with a software controlled generator), CCDSCAM (a plot of CCD temperature for Eos, Atlas and Titan detectors, FIP60 (the same plot for Sapphire, Ruby and Onyx detectors), Cryojet (a plot of temperature and flow rates against frame number) and two further charts for the XrayChiller and CCDChiller.

If data reduction has been carried out several times and saved under different filenames, statistics for each can be observed by locating the appropriate file in the drop-down menu at the bottom of the window.

8.1. Refinalization

If the user wishes to re-process the data, the **Refinalize** button will activate a new window through which many aspects of the data reduction process can be fine tuned (Figure 14). Laue symmetry and outlier rejection can be applied, as well as limits, filters and lattice extinctions.

The data reduction procedure generates a *.rrpprof file, which contains all integrated data. The last step of the data reduction process is finalization, in which a *.hkl file is generated from the *.rrpprof file by applying the Laue symmetry, space group, multiscan absorption correction and frame scaling (both using the ABSPACK module). Data can be Refinalized repeatedly using differing parameters, generating several *.hkl files.

NOTE

The best way of maintaining consistency between all files (*.hkl, *.ins, *.cif) for a given experiment is to use the Refinalization menu to impose any changes

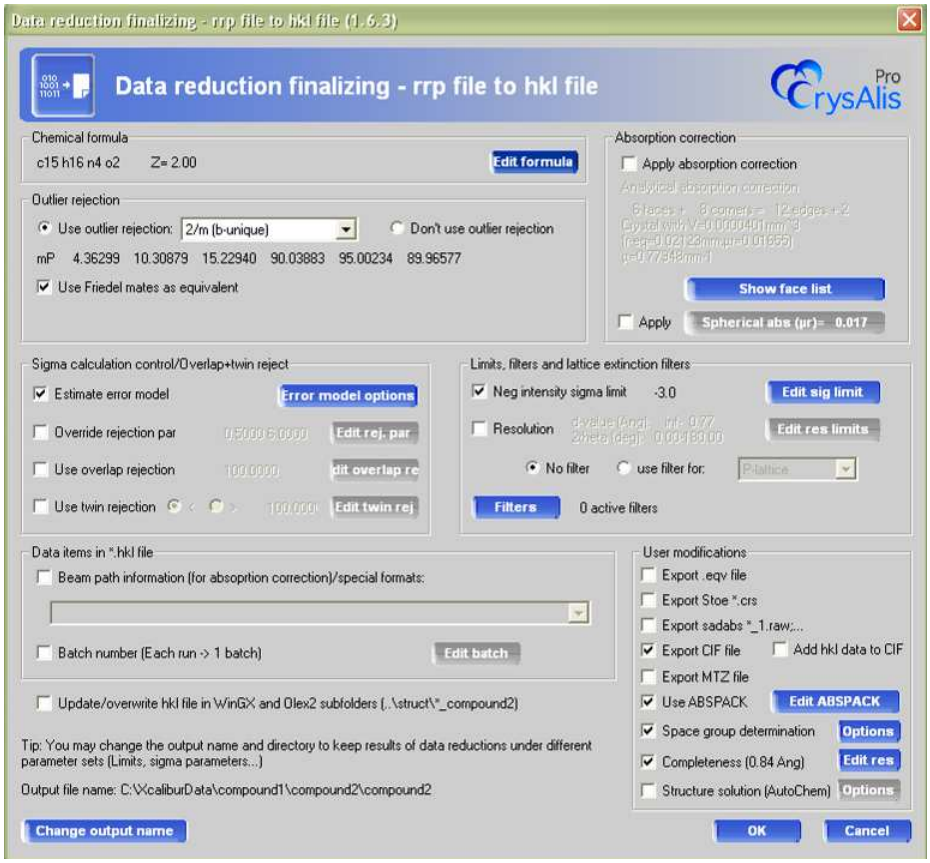


Figure 30 Data reduction refinalisation GUI

8.2. Running GRAL in interactive mode

The space group determination module can be run in interactive mode. This is very helpful in cases where the X-ray data are weak and the automated procedure fails. Interactive GRAL is enabled by opening the Space Group Determination **Options** panel from the Data Reduction Finalizing window (Figure 31) and clicking the appropriate radio button.

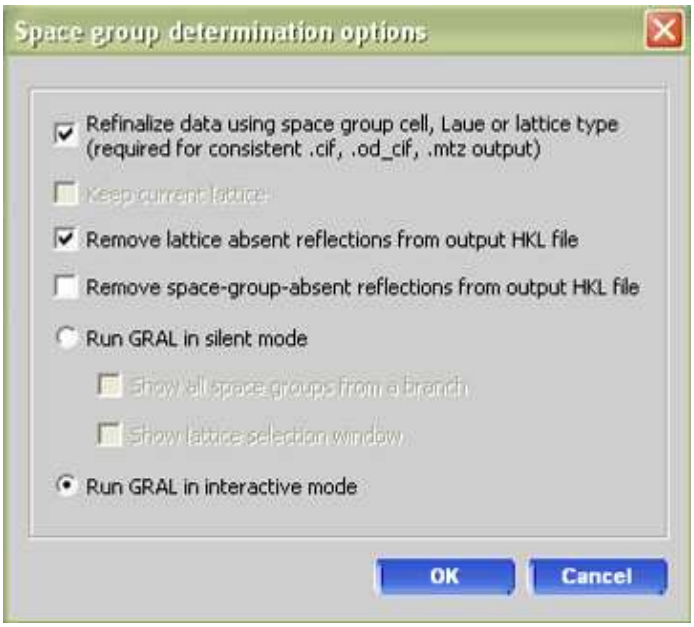


Figure 31 Space group determination options

When interactive GRAL launches, the user is presented with a series of panels showing data relevant to space-group determination. The upper portion of the panel presents the experimental data. Along the bottom are a series of radio buttons. One of these options must be selected before moving on to the next panel. When a panel first appears, one radio button will have been selected automatically by the program. The user must either confirm this choice or select his own choice, based on his interpretation of the experimental data shown in the upper portion of the panel. This process is continued until the space group has been assigned and the unit-cell contents established.

A detailed listing of the content of each panel and possible user responses follows.

1. First the lattice centring is determined. A table of statistics allows the user to check the selection. If the user wishes to choose an alternative then the relevant radio button can be selected by clicking on it and then **Apply** can be clicked to move to the next step.
2. A Niggli reduction is applied to the data and the user is provided the opportunity to apply a transformation matrix to the original cell. Click on **Apply**.
3. The space group power tool provides a list of possible Bravais lattice and highlights in blue the programmes' preferred choice. Clicking on **Apply** will accept the choice, however, any one of the alternatives may be selected by clicking on the option in the text box and then clicking **Apply**.
4. Following Niggli reduction the centring of the cell is re-examined. Click on **Apply**.
5. The data are now examined for acentric or centric E statistics. The user can then choose between Centrosymmetric or Non-centrosymmetric settings by clicking on the relevant radio button and clicking on **Apply**.

6. Space group now examines the systematic absence exceptions (where applicable) and provides a statistical table for user confirmation of its preferred choice of space group. The user can accept the preferred choice (which is highlighted) choice by clicking on **Apply** or select from one of the alternative choices when present.
7. Finally, a *.ins file is prepared for use with structure solution programs. The user can edit the molecular formula and Z number in the relevant boxes. Clicking on **Test** allows the user to check the formula weight, absorption correction μ, density etc against the molecular formula and Z number. The current contents of the *.ins file are displayed on the left hand side of the screen. Clicking on **Finish** creates the *.ins and *.hkl file, and closes the power tool.

8.3. Filtering data

The **Filters** menu enables the user to omit specific runs, frames, intensities, theta values, I/σ values and a variety of other measures from the refinalised data. In the filters menu, click **Add** and then set up a filter using the rejection condition and specific value. The most useful and commonly used filters are for Run, Run-frame, d-value and Rint.

To omit a particular run, simply use the equals (=) condition and choose the run number that you would like to remove. Severe data scaling may be the reason for removing a run and so the run number can simply be read from the absscale plot.

To omit a single frame, use the run-frame filter (= run number frame number).

To omit blocks of frames (perhaps due to icing), use the same filter with two conditions. For example, to omit frames 11 to 20 of run 2, use the condition (> 2 10) but then add a second condition (<= 2 20).

NOTE

Filtering data will inevitably lead to a loss of completeness. For this reason, filters should only be used when absolutely necessary

8.4. Applying absorption corrections

A face indexed absorption correction can be applied (provided the crystal shape has already been determined – see section **8.4**) by ticking the **Apply absorption correction** box in the top right corner of the window. The absorption coefficient (μ) is not always calculated automatically, and so this can be done by setting the formula and Z value in the **Show face list** button. A spherical absorption correction can also be applied by ticking **Apply** next to the **Spherical abs** button and then entering a suitable value for μ*r, where r is the average radius of the crystal.

The transmission factors resulting from the absorption correction are output in the *.cif file generated by refinalization.

8.5. Crystal shape modelling (face indexing)

The **Abs display** button opens the crystal movie window display (provided that a movie of the crystal has been previously recorded). The crystal can be rotated incrementally by pressing the Page Up or Page Down keys.

The crystal shape is defined with respect to both hkl and xyz values. The hkl indices relate to the cell defined at the time of face-indexing. However, as the cell is also defined by standard xyz coordinates, if the cell is changed in any way, the orientation of the model will not change. However, the hkl indices of the faces will change according to the new cell and may no longer be integer values.

Initially, the crystal centre must be defined by right clicking in the appropriate place and choosing **Define center**. Clicking the left mouse button and dragging will drag out two vectors (for principle and mirror faces). The vector needs to be dropped so that it is coincident with a face, with the face perpendicular to the screen. The hkl values for the drawn vector are displayed at the bottom of the window. The face can be added to the list by right clicking on the window and selecting the **Add face** (or add mirror faces) option in the pop-up window. Ideally, natural faces should have natural indices (1 0 0, 0 1 0 etc.). However, it is more likely that faces such as 1 0 10 and 22 0 3 will be observed. These are in fact 0 0 1 and 1 0 0, but due to the errors involved the vectors do not correspond exactly to the natural face.

The user can manually edit the face list as well as control more aspects of the indexing process within the **Faces** tab of the **Crystal movie configuration** window. Once enough faces have been added to define a 3D shape, the shape model will appear superimposed over the movie image. Clicking on a face in the **Faces** tab will highlight it in red on the image, allowing for simple identification and editing.

Refinement of the defined crystal shape can be carried out by clicking on the **Scale3 abs** button in the **Data reduction results** window.

A movie which clearly demonstrates the face indexing process with a worked example is available to download from the Agilent X-ray user forum. This can be accessed at forum.oxford-diffraction.com

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9. Twinning

A twin is formed when two or more crystals of the same material intergrow. The unit cells of the two (or more) components are related to one another by symmetry. The lattices may completely coincide in all three dimensions (merohedral twinning) or in fewer than three dimensions (non-merohedral twinning).

A twin may be suspected for a number of reasons:

- Re-entrant faces on the sample
- Non-uniform polarisation of light when examined by microscope
- Peaks which appear split in the diffraction pattern
- Difficulty with indexing, or a low percentage of indexed peaks
- Difficulty solving and refining the structure to a decent R_1 value
- Two or more clearly visible lattices in the Ewald explorer 3-dimensional lattice viewer

If a twin is suspected before data collection, it is often beneficial to move the detector further back than you would for a standard data collection. This will help to separate the spots from each twin component.

Consider using slightly longer exposure times and collecting higher redundancy than normal to increase the number of data points.

A movie which clearly demonstrates the twin data processing utilities with a worked example is available to download from the Agilent X-ray user forum. This can be accessed at forum.oxford-diffraction.com.

Users will find the movie particularly useful when using the twin data processing tools for the first time.

9.1. Automatic twin lattice finding

Searching for twin lattices is a remarkably simple procedure, to the extent that it is worth exploring the possibility that crystals are twinned whenever problematic data sets are encountered.

To access the automated multiple lattice finding tool, select the **Unit cell finding with options** utility under **Unit cell finding** in the lattice wizard;

1. Select the **Twin / multicrystal** radio button and then choose the number of components to search for. It is often best to look for two lattices at first, and then look for further cells if this seems necessary on further inspection of remaining un-indexed reflections.
2. The 'Force identical lattice for all components' box is ticked automatically. If the routine still finds two very different lattices, the crystal is unlikely to be a twin.
3. Certain crystals will output suspiciously large cells due to the presence of false long axes caused by superimposed twin lattices. In order to prevent this, twin lattice finding can be conducted with added cell axis constraints. The worked example in the twinning movie is one such case where constraints are required in order to find cells of a reasonable size. In most cases, choosing an upper limit of 20-30 Å usually helps. The **Calc** button will bring up a window which will estimate the volume and cell dimensions of the unit cell based on a supplied chemical formula and crystal system.
4. Click on **OK** and the two or more cells output by the cell finding routine will be displayed in the left-hand window. These should be more or less identical. Twin indexation is carried out as part of the automatic process.

5. The cells are automatically stored in the **Twinning - multi-crystal** utility. The relationship between the cells is displayed in this window as a rotation;

   Rot(UB1,UB2) = x deg around a* b* c* (rec) a b c (dir)

6. The relationships between all of up to 4 components are displayed.

7. One of the best methods of assessing the quality of twin lattice finding is visual inspection using the **Ewald explorer** module (See section **9.3** below).

9.2. Manual twin lattice indexation

In the case that the user would like to impose a user defined cell which is not found by the automatic multiple cell finding routine, use the following steps;

1. Once data collection has finished, carry out peak hunting using the peak hunting icon.

2. Find a suitable unit cell using the various cell finding tools (see section **7.1**).

3. The chosen cell will be printed in the left hand window of the lattice wizard. Save the first twin matrix to memory by clicking on the **Twinning - multi-crystals** icon in the lattice wizard.

4. Click on the radio button **Component 1** then select the **Current UB to twin** button. The UB matrix will be printed in the bottom window. Click on **OK**.

5. Click on the **Ewald explorer** icon and radio button **Show wrong** and only the wrongly indexed reflections will be shown.

6. The indexed reflections need to be skipped in order to index the second twin component. Click on the zoom button to zoom out until all reflections are within the screen. Click on the **Drag** radio button and then hold the left mouse button down and drag a box around the reflections. Right click and select **Mark selection skip** and they will turn red. Click on the pull down menu **Flags** and select **Invert used and skipped**. Click on the radio button **Show: all** followed by **OK** and then click on the unit cell finding icon in the lattice wizard.

7. When the 2nd cell has been found, it will be printed in the left hand window of the lattice wizard.

8. Click on the **Twinning - multi-crystals** icon to store the second lattice as component 2. Select **Component 2** and click on the radio button **Current UB to twin**. You now have two twin components saved to memory.

9. Click on **OK** to exit the **UM TWIN** window.

10. Index and refine the two cells by clicking on the arrow to the right of the twinning multi-crystals icon and selecting **Twin indexation**. Make sure the radio buttons **O1 R1 O2** and **R2** are selected then click on **OK**. The cells will be refined and printed in the lattice wizard window. On prompting click **Yes**.

Once the twin components are found, information about them is displayed in the lattice wizard main panel:

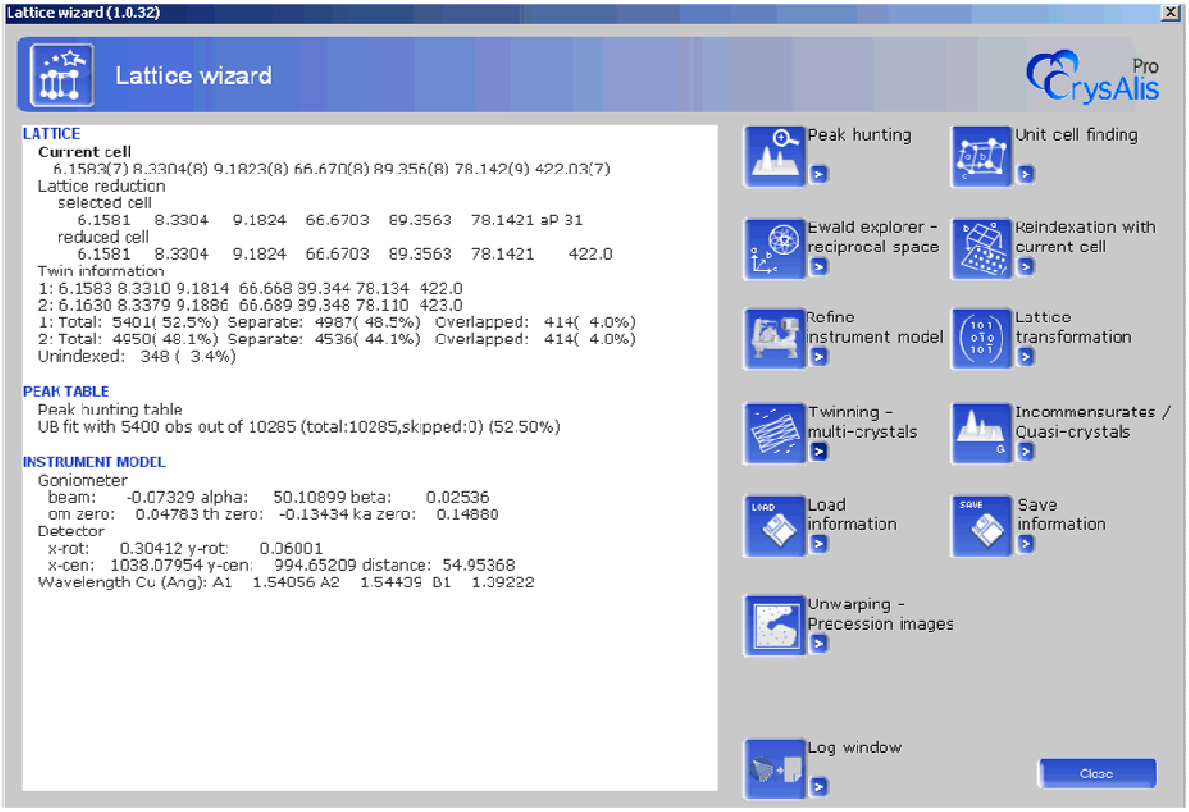

Figure 32 Lattice Wizard panel showing twin information

9.3. Visualising twin components

To visualise twin matrices;

1. Go to the **Ewald explorer** module, click on **Show** in the menu bar and select **Show twin flags and twin lattice** (Figure 15). This will bring up a new window with toggle boxes for up to 4 components. Ticking the **Use twin flag** box will enable the user to toggle each component on or off according to its twin flag. The flags themselves can be seen in the peak table, which is accessible through the small arrow icon next to the Ewald explorer icon.
2. The **Show twin lattice** box enables the user to switch each lattice overlay on and off individually.
3. The **Twin cell – view along** tool means that the sphere of data can be rotated to view along any axis of up to 4 components.
4. If the user is interested to see if the found components account for the majority of the (ordered) reflections, switch on the **Neither** reflections only. By looking at these reflections on their own it is usually easy to see whether or not it is worth attempting to search for further twin components.

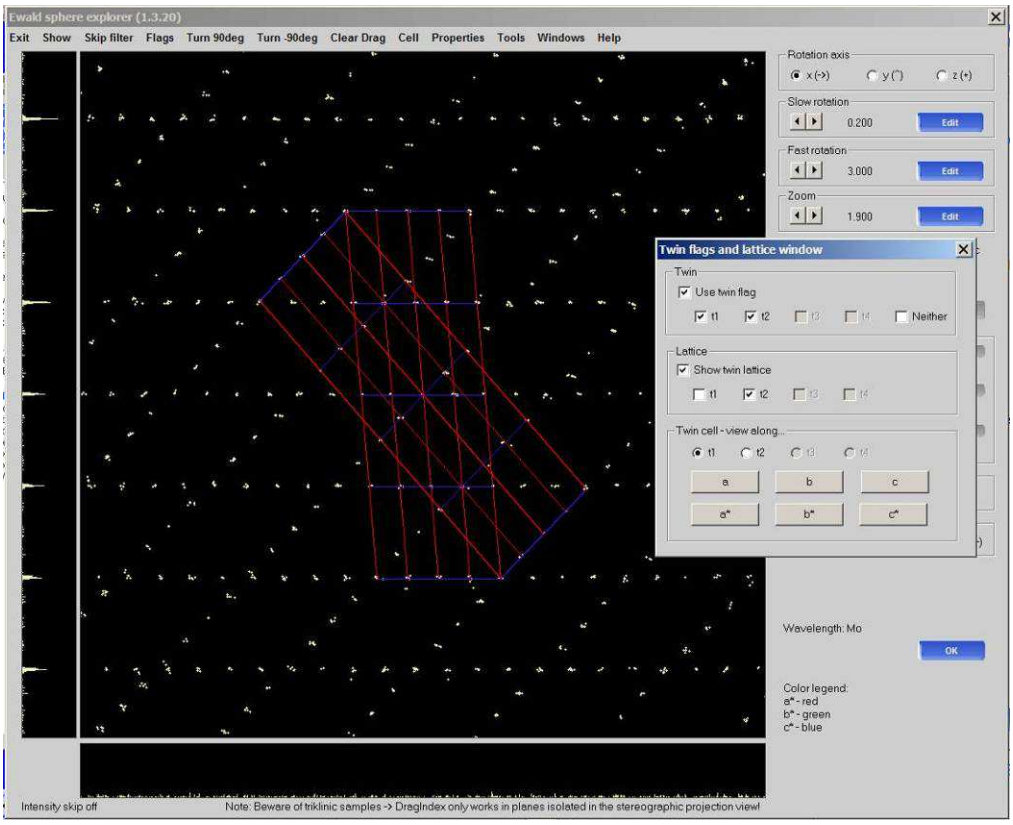

Figure 33 Ewald explorer twin flags and lattice visualisation

9.4. Data reduction of a twinned data set

1. Click on the **START/STOP** button and select **Data reduction with options (Twin)**.
2. The **Twinning – multi-crystal** menu will already be activated with boxes ticked for as many components as have been defined. Switch off components to be integrated as required.
3. Select a suitable follow-model option (see Section 6.5.1). This may prove to be quite important, because the center of mass of individual twin components may not lie at the center of mass of the externally-visible crystal.
4. Continue with data reduction as normal, remembering to **Clear data from previous run** when prompted.
5. Click on **Finish** and data reduction will begin.
6. The frames will be processed repeatedly, once for each twin component.
7. On completion of twin data reduction, a variety of top-level information is displayed in the data reduction tab (Figure 16). This includes the twin ratio, including the number of reflections associated with each component, plus R_{int} and I/sig statistics for both isolated and overlapped reflections (hklf4 and hklf5).

Twin data reduction outputs a single HKLF4 file per twin lattice. Each one is clearly labelled as name_twin1_hklf4.hkl, name_twin2_hklf4.hkl etc. These hklf4 files are standard data files and are to be used for structure solution. The best chances of success come from structure solution using the major component data file, which is usually twin1_hklf4.hkl, however this will depend on which cell was found first and stored as component number 1.

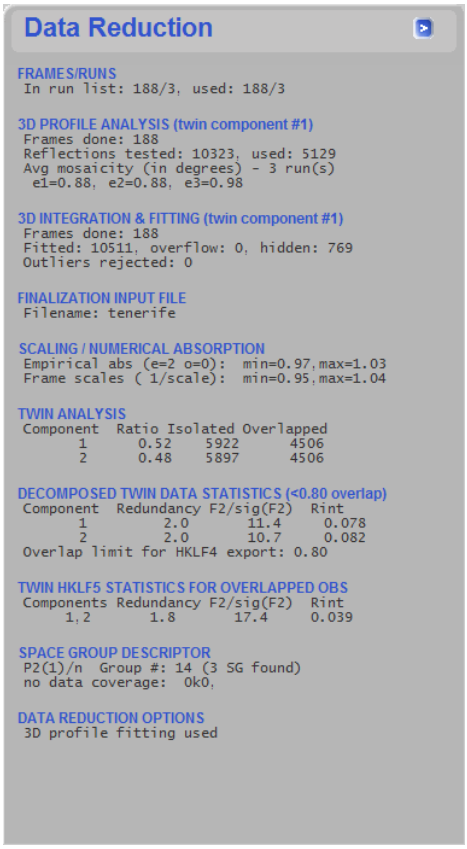

Figure 34 Data Reduction Tab Showing Twin Statistics

In the data reduction tab above there is the line, 'Overlap limit for HKLF4 export: 0.8'. This means that any reflection which is greater than 80% overlapping with another reflection is discarded from the HKLF4 files. This aids structure solution, as some reflections are so severely corrupted that it makes finding a solution almost impossible. The 80% figure works well in most cases, but this can be changed by performing twin finalization (see section **9.5**).

The **Decomposed twin data statistics** give R_{int} values for each component, but these values are sometimes higher than might be expected. This is due to the effect of removing badly overlapped reflections.

The R_{int} value given under **Twin HKLF5 statistics for overlapped observations** gives a very realistic indication as to the success of the twin data reduction. A more detailed output can be found in the **Command Shell**. Here, R_{int} values are displayed for both isolated and overlapped reflections in the HKLF5 output file. This file is labelled name_twin1_hklf5_merged.hkl. As with HKLF4 files, the twin data file incorporates an overlap threshold of 0.8 or 80% (default value). However, no reflections are discarded, but this is the limit at which reflections are either treated separately or as one single entity.

Once a structure solution has been obtained and refinement is at the point where no further improvement can be made, the structure should be refined using the twin data file (HKLF5). In the *.ins file, the HKLF4 command must be changed to HKLF 5. A BASF (batch scale factor) command must also be inserted into the file along with a number which defines the ratio of the twin components. This number will refine, but an initial value is output by CrysAlis^{Pro} and is found as the last item in the command shell. In cases with more than 2 components, more than one BASF number is required. Details of what to include can be found in the command shell.

NOTE

Structure solution cannot be performed using a HKLF5 file. A HKLF4 file containing the information of only a single lattice must be used for this purpose

9.5. Twin data finalization

As with standard data sets, twinned data can be altered by changing the settings in the **Twin finalization** module. This is the 5th power tool, and is only available if twin data reduction has been carried out. All the standard features are available here, including;

- Chemical formula editing
- Laue symmetry and Friedel mates manipulation
- Multiscan absorption and scaling
- Application of face-indexed absorption corrections
- Filters and merging
- Space group determination (including specific control using the space group options window)

One feature here which is unique to twin data finalization is the full overlap threshold setting. The **Full Overlap Threshold** defines the overlap level below which partially-overlapped reflections of twins are partitioned into their component contributions. The parameter can be set to anything between 0 and 1 (with 0.8 as the default value). Changing this threshold may help in cases where no structure solution can be obtained or in cases where refinement with the HKLF5 twin data file yields no improvement. A good example is a case where two lattices are present but look to have little or no impact on one another. In one such case, with 15,000 reflections divided between two components, only 110 of these were overlapped in some way. However, this was enough to cause problems in refinement of the Flack parameter for assignment of absolute configuration. Setting the overlap thresholds to zero and using only the HKLF4 file of the major component for structure refinement fixed the problem completely. This was effectively accepting no overlap of any kind, and so removing any effected reflections from the HKLF4 data file.

Additional features unique to twins are as follows. The **Separate scales for all twin components** option allows for the application of different scaling models for each twin. For some twins this results in a significant improvement of data quality. The **Output multi HKLF4 file** option will attempt to produce a complete data set by combining reflections from the twin components. The reflection intensities are suitably scaled so as to reflect differences in the volume fraction of each twin.

You can also use interactive GRAL in conjunction with twin data. The options **Remove lattice absent reflections from output HKL file** and **Remove space-group-absent reflections from output HKL file** will help assure that you have more complete data in your hklf4 file, provided your sample has space group extinctions that are detectable.

NOTE

It is often a good idea to reconstruct some precession images to directly observe the extent of the overlap between twinned reflections. This is also a fantastic teaching aid for presentations focussed on twinned data sets

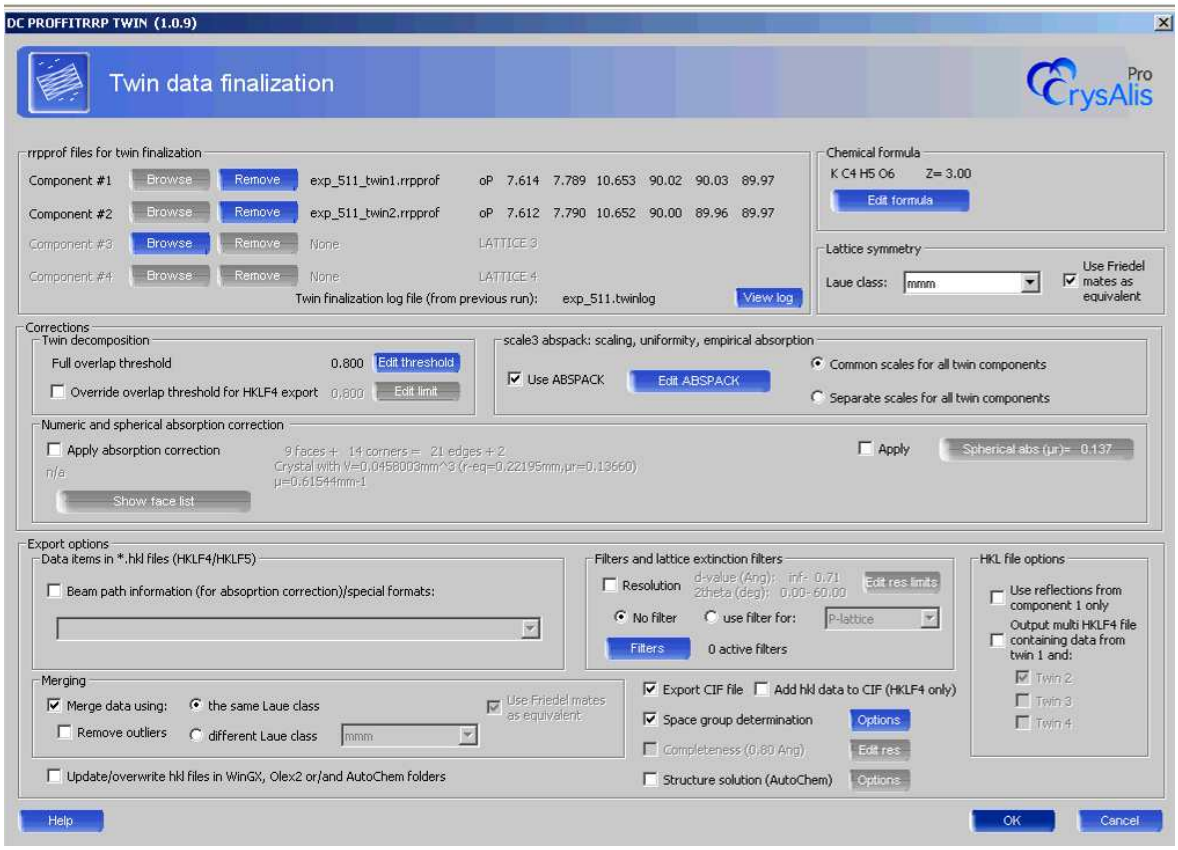

Figure 35 Twin data finalization panel

9.6. Indexing problem twins

If the automatic algorithms fail to index the twinned compound, try to index it manually using the drag index feature described in the Ewald module. To help you, the stereographic projection provides a clear way of finding lattice planes.

Rotate the stereo projection diagram around using the arrow keys until a lattice line is aligned vertically or horizontally. A maximum will appear in the histogram bars at the bottom and far left hand side of the image. If you deselect the stereographic projection radio button, the lattice lines should be lined up and the drag index function should be easier to use.

10. Incommensurates

An incommensurate or modulated structure derives from data where there is periodic distortion of the atomic positions (modulation) and/or of the occupation probability of atoms (density modulation). This gives rise to satellite reflections present in addition to the main lattice. Satellite reflections are often of very weak intensity and so are not always clearly observed. The best chance of observing as many satellite reflections as possible is to collect data with a reduced scan width (~0.5°) and with a significant exposure time. Collecting data of increased redundancy is also beneficial.

A movie demonstrating some of the incommensurate data processing utilities with a worked example is available to download from the Agilent X-ray user forum. This can be accessed at forum.oxford-diffraction.com.

Users will find the movie particularly useful when using incommensurate tools for the first time.

Successfully identifying the parent lattice in an incommensurate crystal is often the key to treating incommensurate data sets. Finding the parent/main unit cell can be as simple as finding a standard cell, but on occasions it can be quite difficult. Structures are often highly symmetrical and so many different cell and space group combinations are possible (the added presence of satellite reflections makes this even more difficult).

One simple technique for identification of the parent lattice is to filter the reflections based on intensity in order to filter out the weaker satellites. This can be done in the **Ewald explorer** module;

1. Use the **Skip filter** and select **Intensity...**
2. Tick the **Remove weak reflections** box and raise the threshold. The weak reflections will gradually disappear.
3. To skip the removed peaks, choose **Mark invisible skip** in the **Flags** menu. The skipped flag will be placed next to the reflections concerned I the peak table and they will not be used in any unit cell determination.
4. Attempt to find a suitable unit cell with a high % fit, accounting for the maximum number of unskipped reflections.

The **Gnomonic** view in the **Ewald Explorer** module combined with the **Drag indexing** function can also be used when it is difficult to distinguish between different cells.

Initially, perform a standard data reduction and attempt to solve and refine the structure as normal. This will be the average structure, with no treatment of modulation taken into account.

Incommensurate data sets with q vectors in up to 3 directions can be handled. Samples that are both incommensurate and twinned can also be dealt with. The best method for searching for potential satellite peaks is by using the **q-vector overlay** tool in the **Ewald Explorer** module.

1. In the menu bar, click **Windows** and select **Show q-vector overlay window** (Figure 17).
2. Up to 3 q-vectors can be defined. To begin, click on **Set overlay for q1** and either **Edit** the a b and c coordinates (with max m/order as 1 to start) or use the **Interactive adjustment** to move the purple overlay spots in the Ewald image.
3. The q-vector should be defined so that the overlay spots line up well with what are considered to be satellite reflections.

4. Once an initial overlay has been set, drop back to the lattice wizard and select the **Incommensurates/Quasi-crystals** icon. The order (m max), hkl integer tolerance (crithkl) and q-vector coordinates from the defined overlay are already input, so simply click on **OK**.

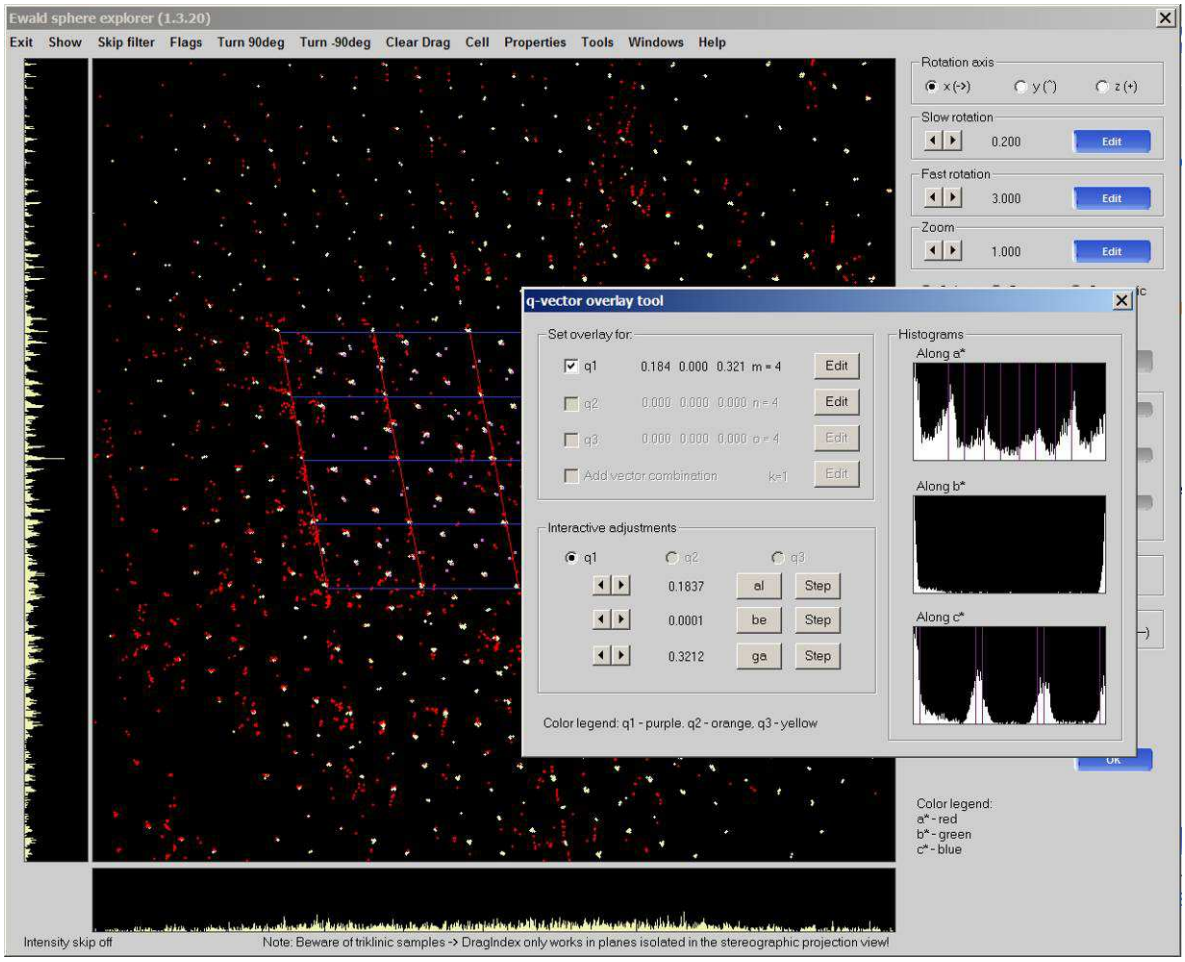

Figure 36 Ewald explorer with q-vector overlay tool

The values of the q-vector coordinates are refined and output information is available under the peak table menu on the left of the lattice wizard window. The key points to note are;

a. Has the % of indexed reflections increased?

b. How many **Main reflections**, **q1 satellites** are there?

The second, third, fourth *etc* orders can now also be investigated by returning to the q-vector overlay tool and raising the value of m max for the q1 satellites. Use visual inspection of the overlay spots as well as the histograms on the right of the window to decide whether or not the specified order makes sense.

1. Raise the value of m max by a single integer (1 to 2).
2. Return to the lattice wizard and refine the q-vector values in the same way as before.
3. Look to see if the % of indexed reflections has increased.

4. Observe the **By order** output to see how many 2nd order satellites are present.
5. If this value is zero, 2nd order satellites are probably not present. Reset the value of mmax to 1 and refine again.
6. However, if 2nd order satellites are present, use the same approach to look for 3rd then 4th order satellites etc.

Crystals with more than a single q-vector are relatively rare. Samples with three q-vectors (3-d quasi-crystals) are extremely rare and so in most cases single q-vectors (possibly with a satellite order higher than 1) are most worthy of pursuit in the first instance.

Once a suitable peak table has been obtained, this can be saved specifically by choosing **Save specific peak table only** under the **Save information button**.

The data must then be integrated by using **Data reduction with options** under the **START/STOP** menu.

1. On the first screen, check that the correct q-vector and order are already defined for **Single q-vector** cases.
2. In more specialist cases, select **Other (reduction list)** and **Generate** an hkl list.
3. The data reduction module will output an hklm file for single q-vector cases (hklmn for 2-d and hklmno for 3-d). Other highly specialist tools are also available in the **Generate** window.
4. Progress through the **Data reduction with options** choices as normal, giving the output a suitable systematic name.
5. Click **Finish**.
6. All output files are compatible with Jana (see section **4.4.15**), for modulated structure refinement.

11. External Detector Frame Formats

11.1. Dectris

CrysAlis^{Pro} now supports processing of Dectris detector frames. At the moment only the detector for Swiss Light Source instrument 6M is implemented. The detector gaps of the Dectris detector are automatically handled.

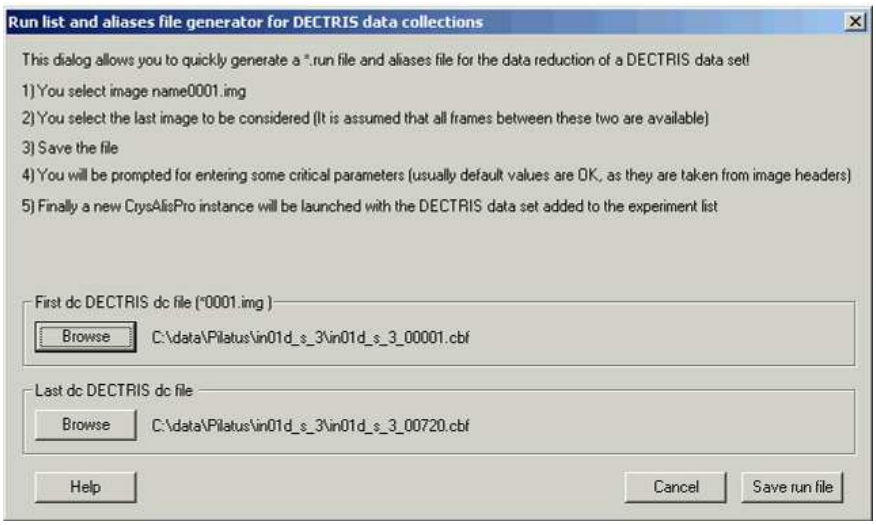

Figure 37 Run list generator for Dectris

11.2. Rigaku

CrysAlis^{Pro} now supports processing of Rigaku d*trek image format frames.

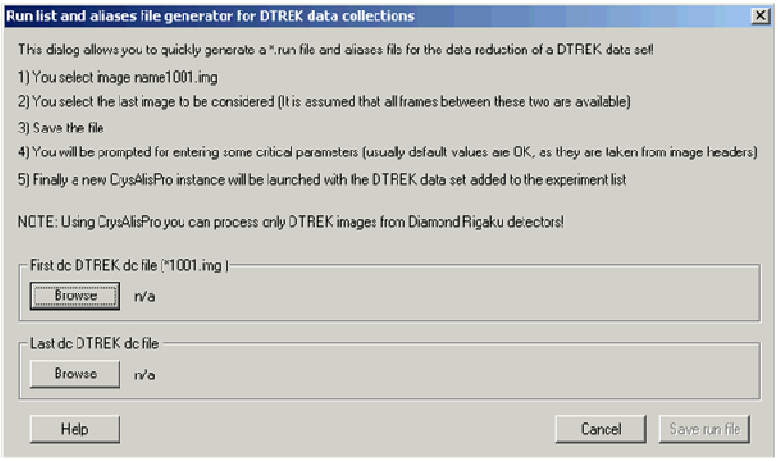

Figure 38 Run list generator for d*trek image format

11.3. SAXI

Data from Bruker AXS-saxi formats can also be processed with CrysAlis[Pro]:

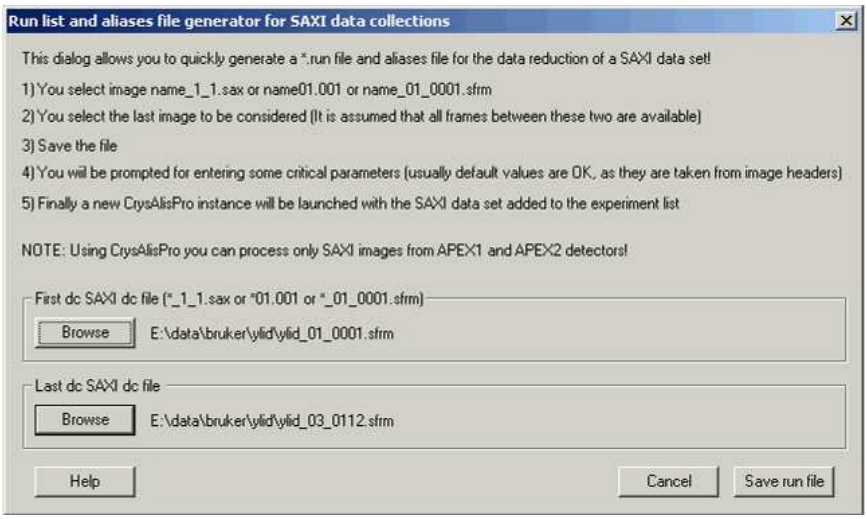

Figure 39 Run list generator for SAXI

11.4. MAR

Data from MAR image plates and some MAR CCD detectors (e.g., MARCCD 165) may be processed with CrysAlis[Pro]:

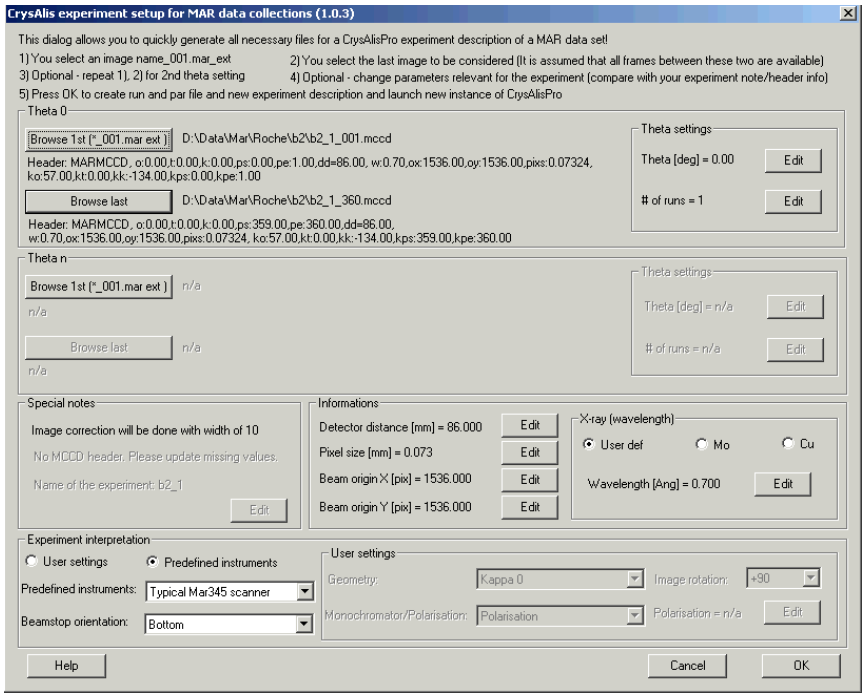

Figure 40 Run list generator for MAR

Appendix

Directory Structure

The CrysAlis^{Pro} program system is installed in the following directory structure;

Windows program files directory (typically C:\)

Xcalibur root (typically C:\Xcalibur)

Corrections directory (\corrections)

Site directory (\sitename)

*.ffi, *.par, *.ccd and *.geo correction files. These files describe the machine setup

Log directory (\log)

*.log. This directory contains the applications error logs, which are important to trace application errors.

Macro directory (\macro)

*.mac. This directory contains the CrysAlis macros for task automation.

Applications root (\CrysAlisXXX)

*.exe, *.dll, *.vxd/*.sys. This directory contains the CrysAlis executables and drivers.

Darkimages directory (\darkimages)

*.img. This directory contains the CrysAlis dark images, which are acquired during a CrysAlis CCD session.

Initialisation and user defined setup files directory (\CrysAlisINI)

This directory contains initialisation , user defined setup files for CrysAlis, ODBench and ODShell programs.

ODShel software directory (\ODshelEXE)

This is an executable folder which contains the ODShel software. This is a shell program for structure solution and refinement.

A typical experiment folder will have the following directory structure;

Experiment root (C:\Xcaliburdata\expname)

This directory contains important experiment definition files such as *.par, *.run, *.ffiinffit, *.ffi, *.tab, *.rrpprof, *.ccd, *.geo, *.gon*.sum, *.cif, *.ins, *.hkl etc. all of which are required in order for the experiment to open and function properly.

Expinfo directory (\expinfo)

This contains *.ini files with information regarding some of the unique settings for this experiment.

Frames directory

Contains all the frames in *.img format.

Log directory

Contains logs for CCD, RED and USER threads in *.txt format.

Movie directory

This contains *.jpg images collected as a rotated movie of the crystal.

Plotsdc directory

*.dat files contain information logged from the various hardware components of the system.

Plotsred directory

*.dat files contain information on scaling, R_{int} and other integrated data output functions.

Struct directory

This is set aside for storing structure solution and refinement files.

Tmp directory

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